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EXAMINER	
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ART UNIT	**PAPER NUMBER**
1637	

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Please find below and/or attached an Office communication concerning this application or proceeding.

Office Action Summary	Application No.	Applicant(s)	
	10/521,721	FREBOURG ET AL.	
	Examiner	Art Unit	
	Angela Bertagna	1637	

-- The MAILING DATE of this communication appears on the cover sheet with the correspondence address --

Period for Reply

A SHORTENED STATUTORY PERIOD FOR REPLY IS SET TO EXPIRE 3 MONTH(S) OR THIRTY (30) DAYS, WHICHEVER IS LONGER, FROM THE MAILING DATE OF THIS COMMUNICATION.

- Extensions of time may be available under the provisions of 37 CFR 1.136(a). In no event, however, may a reply be timely filed after SIX (6) MONTHS from the mailing date of this communication.
- If NO period for reply is specified above, the maximum statutory period will apply and will expire SIX (6) MONTHS from the mailing date of this communication.
- Failure to reply within the set or extended period for reply will, by statute, cause the application to become ABANDONED (35 U.S.C. § 133). Any reply received by the Office later than three months after the mailing date of this communication, even if timely filed, may reduce any earned patent term adjustment. See 37 CFR 1.704(b).

Status

1)☒ Responsive to communication(s) filed on *23 August 2006*.

2a)☐ This action is **FINAL**. 2b)☒ This action is non-final.

3)☐ Since this application is in condition for allowance except for formal matters, prosecution as to the merits is closed in accordance with the practice under *Ex parte Quayle*, 1935 C.D. 11, 453 O.G. 213.

Disposition of Claims

4)☒ Claim(s) *1-40* is/are pending in the application.

4a) Of the above claim(s) *1-25 and 40* is/are withdrawn from consideration.

5)☐ Claim(s) _____ is/are allowed.

6)☒ Claim(s) *26-39* is/are rejected.

7)☒ Claim(s) *26-39* is/are objected to.

8)☐ Claim(s) _____ are subject to restriction and/or election requirement.

Application Papers

9)☒ The specification is objected to by the Examiner.

10)☒ The drawing(s) filed on *18 April 2005* is/are: a)☒ accepted or b)☐ objected to by the Examiner.

Applicant may not request that any objection to the drawing(s) be held in abeyance. See 37 CFR 1.85(a).

Replacement drawing sheet(s) including the correction is required if the drawing(s) is objected to. See 37 CFR 1.121(d).

11)☐ The oath or declaration is objected to by the Examiner. Note the attached Office Action or form PTO-152.

Priority under 35 U.S.C. § 119

12)☒ Acknowledgment is made of a claim for foreign priority under 35 U.S.C. § 119(a)-(d) or (f).

a)☒ All b)☐ Some * c)☐ None of:

1.☒ Certified copies of the priority documents have been received.

2.☐ Certified copies of the priority documents have been received in Application No. _____.

3.☐ Copies of the certified copies of the priority documents have been received in this National Stage application from the International Bureau (PCT Rule 17.2(a)).

* See the attached detailed Office action for a list of the certified copies not received.

Attachment(s)

1) ☒ Notice of References Cited (PTO-892)
2) ☐ Notice of Draftsperson's Patent Drawing Review (PTO-948)
3) ☒ Information Disclosure Statement(s) (PTO/SB/08) Paper No(s)/Mail Date *1/19/05; 2/22/05*.
4) ☐ Interview Summary (PTO-413) Paper No(s)/Mail Date. _____.
5) ☐ Notice of Informal Patent Application
6) ☐ Other: Exhibit A, Exhibit B

DETAILED ACTION

Election/Restrictions

1. Applicant's election with traverse of Group III, claims 26-39 and SEQ ID Nos: 3 and 4, in the reply filed on August 23, 2006 is acknowledged. Applicant has traversed the finding of lack of unity between Groups I-III and also the requirement to select a specific primer pair for examination. Regarding the lack of unity between Groups I-III, Applicant argues that, contrary to the Restriction Requirement mailed May 23, 2006, the prior art of Wang et al. (WO 99/58721) does not teach all of the limitations of the instant claim 9, and therefore, the claims possess a special technical feature linking them over the prior art. This argument was not found persuasive, because although Applicant is correct that Wang does not teach all of the elements of claim 9, the prior art of Duponchel et al. (Human Mutation (2001) 17: 61-70) teaches all of the elements of claims 9 and 26. Therefore, since the prior art of Duponchel anticipates the instant claims 9 and 26, the claims lack a special technical feature linking them over the prior art, and a lack of unity requirement is proper. Applicant also traversed the requirement to select a specific primer pair for examination on the grounds that the Office Action did not demonstrate that the claimed sequences are independent inventions. Applicant cites the fact that the sequences have a common function as evidence that they do not constitute separate inventions. This argument was also not found persuasive, because each different primer pair consists of two different nucleic acid sequences that do not share a common structural core (i.e. they are not homologous variants or degenerates). Also, since each primer pair is designed to amplify a different target sequence, the primers do not share a common function. Therefore, as discussed previously, each primer pair constitutes an independent invention, subject to a lack of unity requirement.

The requirement is still deemed proper and is therefore made FINAL.

Claims 1-25 and 40 and also the non-elected sequences are withdrawn from further consideration pursuant to 37 CFR 1.142(b), as being drawn to a nonelected invention, there being no allowable generic or linking claim. Applicant timely traversed the restriction (election) requirement in the reply filed on August 23, 2006.

Applicant is reminded that upon the cancellation of claims to a non-elected invention, the inventorship must be amended in compliance with 37 CFR 1.48(b) if one or more of the currently named inventors is no longer an inventor of at least one claim remaining in the application. Any amendment of inventorship must be accompanied by a request under 37 CFR 1.48(b) and by the fee required under 37 CFR 1.17(i).

Priority

2. Receipt is acknowledged of papers submitted under 35 U.S.C. 119(a)-(d), which papers have been placed of record in the file.

Information Disclosure Statement

3. It is noted that the IDS filed February 22, 2005 lists several references that are also cited on the IDS filed January 9, 2005. The duplicate citations have been lined through on the IDS filed February 22, 2005 and considered on the IDS filed January 9, 2005.

Specification

4. The disclosure is objected to because of the following informalities:

(1) The disclosure is objected to because it contains an embedded hyperlink and/or other form of browser-executable code. Applicant is required to delete the embedded hyperlink and/or other form of browser-executable code. See MPEP § 608.01. The embedded hyperlinks appear on pages 20 and 25.

(2) The "Brief Description of the Drawings" heading is missing

(3) Tables 1 and 3 list nucleic acid sequences greater than 10 nucleotides in length that are not identified by the appropriate SEQ ID NO.

Appropriate correction is required.

Claim Objections

5. Claims 26-39 are objected to because of the following informalities: These claims depend from non-elected claim (claim 9). Incorporation of the limitations of claim 9 into elected claim 26 would correct this problem. Appropriate correction is required.

Claim Rejections - 35 USC § 112 - 1st paragraph (Scope of Enablement)

6. The following is a quotation of the first paragraph of 35 U.S.C. 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

Claim 39 is rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the enablement requirement. The claim(s) contains subject matter which was not described in the

specification in such a way as to enable one skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or use the invention.

Factors to be considered in determining whether a disclosure meets the enablement requirement of 35 USC 112, first paragraph, have been described by the court in *In re Wands*, 8 USPQ2d 1400 (CA FC 1988). *Wands* states at page 1404,

> "Factors to be considered in determining whether a disclosure would require undue experimentation have been summarized by the board in Ex parte Forman. They include (1) the quantity of experimentation necessary, (2) the amount of direction or guidance presented, (3) the presence or absence of working examples, (4) the nature of the invention, (5) the state of the prior art, (6) the relative skill of those in the art, (7) the predictability or unpredictability of the art, and (8) the breadth of the claims."

Enablement issues

The fundamental enablement problem with the instant claim 39 is that the claim is directed to a method of diagnosing any genetic disease or predicting an individual's propensity to develop any genetic disease based solely on a determination of the presence or absence of an unspecified genomic rearrangement, whereas the disclosure does not demonstrate a single example of a positive diagnosis of any disease based solely on the genetic rearrangement screening results.

The nature of the invention

Claim 39 is drawn to a method of diagnosing a genetic disease or predicting an individual's propensity to develop a genetic disease based solely on a determination of the presence or absence of a genomic rearrangement. The invention is in a class of invention that the CAFC has characterized as "the unpredictable arts such as chemistry and biology." Mycogen Plant Sci., Inc. v. Monsanto Co., 243 F.3d 1316, 1330 (Fed. Cir. 2001).

The breadth of the claims

Claim 39 is broadly drawn to a method of diagnosing *any* genetic disease or predicting an individual's propensity to developing *any* genetic disease based solely on a determination of the presence or absence of an unspecified genomic rearrangement. The specification asserts that the method may be applied to a number of different diseases including: breast cancer, hereditary non-polyposis colorectal cancer (HNPCC), infantile spinal muscular atrophy, schizophrenia, mental retardation, Von Hippel Lindau cancer syndrome, multiple endocrine neoplasia, neurofibromatosis, retinoblastoma, Peutz-Jeghers syndrome, Sotos syndrome, Smith Magenis syndrome, and beta thalassemias (page 50). The range of diseases and conditions encompassed by claim 39 inherently possess radically different etiologies and symptoms and in many cases have no relationship to each other whatsoever. In short, the claims are extremely broad in scope, covering diagnosis of an immense number of diseases based solely on detection of an unspecified genomic rearrangement, whereas the specification fails to positively diagnose even a single disease or condition based solely on the presence or absence of a genomic rearrangement determined using the method of the invention.

Quantity of Experimentation

The quantity of experimentation in this area is immense, since there is complete variability as to whether or not the observation of a particular genomic rearrangement is sufficient to positively diagnose the individual in the absence of other diagnostic criteria. It would require significant study and experimentation including trials with hundreds of patients to determine that the presence or absence of even a single genomic rearrangement is capable of reliably functioning alone as the diagnostic indicator for a particular disease. This would be an inventive, unpredictable and difficult undertaking in itself, and the efficacy of any of the observed rearrangements as a diagnostic marker for any particular disease would need to be demonstrated in a variety of patients with a statistically significant result. This would require years of inventive effort, with each of the many intervening steps, upon effective reduction to practice, not providing any guarantee of success in the succeeding steps.

Wacholder et al. (J. Natl. Cancer Institute (2004) 96(6): 434-442) notes that in studies of the association of mutations with specific diseases larger studies with 1500 participants have significantly more statistical power than smaller studies (see page 435). So, the quantity of experimentation factor supports the conclusion that a large quantity of experimentation, with the use of many hundreds, perhaps even thousands, of patient samples would be necessary to demonstrate an association between even one of genomic rearrangements and a specific disease disclosed by Applicant. To cover all possible disclosed genomic rearrangements tens of thousands of patient samples would be necessary, and to cover any fraction of the range of the disclosed diseases and conditions, hundreds of thousands of separate patients and the associated analyses would be required. This is a very large amount of experimentation.

State of the Prior Art

The art teaches that it is entirely unpredictable whether or not genomic rearrangements are associated with a given disease. For example, Charbonnier et al. (Cancer Research (2001) 62: 848-853) analyzed 92 families fulfilling (at least partially) the Amsterdam criteria for hereditary nonpolyposis colorectal cancer (HNPCC) for genomic rearrangements in the MSH2, MLH1, and MSH6 genes (see abstract and Materials & Methods). Although thirteen rearrangements were identified in the MSH2 gene, no genomic rearrangements were found in the MLH1 and MSH6 genes (see abstract). Similarly, Saugier-Veber analyzed twelve genes involved in mental retardation for genomic rearrangements, but only identified three rearrangements (see abstract). These studies highlight the fact that it is highly unpredictable whether or not genomic rearrangements are found in genes suspected to be associated with specific diseases.

Furthermore, the art is replete with evidence that gene association studies are typically wrong. In fact, Lucentini et al (The Scientist (2004) Vol 18) titled his article "Gene Association Studies Typically Wrong" and states "Two recent studies found that typically, when a finding is first published linking a given gene with a complex disease, there is only roughly a one-third chance that studies will reliably confirm the finding (see page 2 of printout)." This is consistent with the teaching of Wacholder et al (J. Natl. Cancer Institute (2004) 96(6): 434-442) who notes, "Too many reports of associations between genetic variants and common cancer sites and other complex diseases are false positives" (see abstract). Ioannidis (Nature genetics (2001) 29:306-309) further supports this conclusion in pointing out the heterogeneity of results among different studies of genetic polymorphisms (see abstract, for example).

Therefore, the art suggests that the detection of genomic rearrangements is not always correlated with positive disease diagnosis, but rather must be combined with additional test results. The art also suggests that many reported associations between variant polynucleotides may be incorrect, thereby providing support for the conclusion that it is entirely unpredictable whether a given variant gene will function in a diagnostic capacity for a given disease.

Working Examples

The specification contains several working examples were the claimed method is applied to detection of genomic rearrangements (see pages 59-76). However, the nucleic acid samples used in these examples were isolated from patients already diagnosed with a specific disease (colorectal cancer, schizophrenia, etc) rather than an undiagnosed sample, and no indication is given, either in the examples, or elsewhere in the specification that the method outlined in the working examples could be used to positively diagnose even one disease in the absence of additional tests.

Guidance in the Specification

The specification teaches generally that genomic rearrangements may be used to diagnose a number of diseases (page 50). In some instances, specific regions where rearrangements may occur are taught (see page 51 and the working examples, which targeted specific regions as potentially containing genomic rearrangments). However, the specification fails to teach that a positive diagnosis of even one of the disclosed diseases is possible based solely on the presence of a genomic rearrangement. As noted above, the working examples were

conducted using samples from patients already diagnosed with the particular disesase, and therefore, function as a means of assay validation rather than a means of diagnosing subjects with unknown disease status. Finally, the specification provides no guidance on methods or techniques to demonstrate an association between any specific disease and any specific genomic rearrangement. The specification even fails to provide any discussion or description of the scientific steps necessary to provide evidence that would associate a particular gene or variant thereof with a specific disorder from the extensive list of different types diseases and conditions.

Level of Skill in the Art

The level of skill in the art is deemed to be high.

Conclusion

In the instant case, as discussed above, the level of unpredictability in the association of any genomic rearrangement and any particular disease, where there is no teaching in the specification or art that any specific gene or gene variant is sufficient alone to diagnose any disease, in concert with the teaching that many published association studies are simply wrong supports a finding of undue experimentation. The specification provides the ordinary practitioner with no written description or guidance that leads to a reliable method of associating any specific differentially expressed or variant gene with any disease state. Furthermore, the specification does not provide guidance to overcome art-recognized problems in the association of mutations with diseases as shown by Lucentini, Wacholder, and Ionnadis. Finally, the quantity of experimentation is immense. Thus, given the broad claims to the diagnosis of a large

number of pathologies based on the detection of any genomic rearrangement in an art whose nature is identified as unpredictable, the unpredictability of that art, the large quantity of research required to define these unpredictable variables, the lack of guidance provided in the specification, the absence of any working examples and the negative teachings in the prior art balanced only against the high skill level in the art, the inevitable conclusion is that it would require undue experimentation for one of skill in the art to perform the method of the claims as broadly written.

Claim Rejections - 35 USC § 112 – 1st paragraph (Written Description)

7. The following is a quotation of the first paragraph of 35 U.S.C. 112:

> The specification shall contain a written description of the invention, and of the manner and process of making and using it, in such full, clear, concise, and exact terms as to enable any person skilled in the art to which it pertains, or with which it is most nearly connected, to make and use the same and shall set forth the best mode contemplated by the inventor of carrying out his invention.

Claims 26-39 are rejected under 35 U.S.C. 112, first paragraph, as failing to comply with the written description requirement. The claim(s) contains subject matter which was not described in the specification in such a way as to reasonably convey to one skilled in the relevant art that the inventor(s), at the time the application was filed, had possession of the claimed invention.

Claims 26-39 are directed to a multiplex amplification method utilizing composite primers claimed in terms of their thermodynamic properties (see claim 9). Specifically, the primers must include a 5' tag sequence that does not hybridize to the target nucleic acid, and furthermore, this tag must increase the melting temperature of the primer 10-15C (part d of claim 9). Also, the primers must not hybridize to themselves or other primers in the reaction mixture

with a free energy greater than 14 kcal/mole as determined by "Primer Premier" software version 5.0 (part e of claim 9).

Applicant's specification teaches general rules for calculating free energies of hybridization based on nearest neighbor interactions (pages 19-22), but does not describe how the free energies required by claim were calculated other than to cite the aforementioned software program. Applicant neither describes the control parameters used when calculating free energies using the Primer Premier software nor presents any sort of output demonstrating that the disclosed composite primers possess the required free energies. Moreover, the Primer Premier software may change during the pendency of the application or any patent issued thereupon. For example, the algorithm used to calculate thermodynamic properties may change while the software retains the version 5.0 designation. Such changes would make it virtually impossible for an ordinary practitioner to know whether or not a particular primer pair infringed upon the instant claims. Finally, since neither the software nor the specific algorithm used to calculate the thermodynamic properties of the oligonucleotides is freely available, an ordinary practitioner would be required to purchase potentially superfluous software in order to determine if the instant claims have been infringed upon. In short, it is entirely unclear whether or not the claimed primers possess the required free energies since Applicant has not presented the appropriate data or taught one of ordinary skill how to calculate the free energies using the software. Therefore, it must be concluded that Applicant did not have possession of the claimed invention at the time of filing.

Claim Rejections - 35 USC § 112 – 2nd paragraph

8. The following is a quotation of the second paragraph of 35 U.S.C. 112:

> The specification shall conclude with one or more claims particularly pointing out and distinctly claiming the subject matter which the applicant regards as his invention.

Claims 26-39 are rejected under 35 U.S.C. 112, second paragraph, as being indefinite for failing to particularly point out and distinctly claim the subject matter which applicant regards as the invention.

Claims 26-39 are indefinite, because use of the phrase "characterized in that" in claims 26-39 causes the scope of the claims to be unclear. Substitution of this phrase with language such as "comprising" or "wherein" should correct this problem.

Claims 26-39 are further indefinite, because these claims recite phrases such as "a candidate region…is selected" and "the intensity of amplification…is measured" (see steps a and d of claim 32, for example). While minute details are not required in method claims, at least the basic steps must be recited in a positive, active fashion. See Ex parte Erlich, 3 USPQ2d, p. 1011 (Bd. Pat. App. Int. 1986).

Claims 26-39 are further indefinite, because claim 26 requires the use of the primers defined in any of claims 9-18 in the multiplex amplification method. Claim 9 contains the trademark/trade name "Primer Premier" software. Where a trademark or trade name is used in a claim as a limitation to identify or describe a particular material or product, the claim does not comply with the requirements of 35 U.S.C. 112, second paragraph. See *Ex parte Simpson*, 218 USPQ 1020 (Bd. App. 1982). The claim scope is uncertain since the trademark or trade name cannot be used properly to identify any particular material or product. A trademark or trade name is used to identify a source of goods, and not the goods themselves. Thus, a trademark or

trade name does not identify or describe the goods associated with the trademark or trade name. In the present case, the trademark/trade name is used to identify/describe software for calculating thermodynamic properties of primers and, accordingly, the identification/description is indefinite.

Claim 28 is further indefinite, because it recites a broad and narrow range in the same claim. A broad range or limitation together with a narrow range or limitation that falls within the broad range or limitation (in the same claim) is considered indefinite, since the resulting claim does not clearly set forth the metes and bounds of the patent protection desired. See MPEP § 2173.05(c). Note the explanation given by the Board of Patent Appeals and Interferences in *Ex parte Wu*, 10 USPQ2d 2031, 2033 (Bd. Pat. App. & Inter. 1989), as to where broad language is followed by "such as" and then narrow language. The Board stated that this can render a claim indefinite by raising a question or doubt as to whether the feature introduced by such language is (a) merely exemplary of the remainder of the claim, and therefore not required, or (b) a required feature of the claims. Note also, for example, the decisions of *Ex parte Steigewald*, 131 USPQ 74 (Bd. App. 1961); *Ex parte Hall*, 83 USPQ 38 (Bd. App. 1948); and *Ex parte Hasche*, 86 USPQ 481 (Bd. App. 1949). In the present instance, claim 28 recites the broad recitation 90 to 500 bp, and the claim also recites 90 to 300 bp, which is the narrower statement of the range/limitation.

Regarding claim 29, the phrase "such as" renders the claim indefinite because it is unclear whether the limitations following the phrase are part of the claimed invention. See MPEP § 2173.05(d).

Claim Rejections - 35 USC § 102

9. The following is a quotation of the appropriate paragraphs of 35 U.S.C. 102 that form the basis for the rejections under this section made in this Office action:

> A person shall be entitled to a patent unless –
>
> (a) the invention was known or used by others in this country, or patented or described in a printed publication in this or a foreign country, before the invention thereof by the applicant for a patent.
>
> (b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States.

10. Claims 26-33 and 35-38 are rejected under 35 U.S.C. 102(a) as being anticpated by Casilli et al. (Human Mutation (September 2002) 20: 218-226; cited on IDS).

Applicant cannot rely upon the foreign priority papers to overcome this rejection because a translation of said papers has not been made of record in accordance with 37 CFR 1.55. See MPEP § 201.15.

Regarding claim 26, Casilli teaches a method for amplifying in multiplex a plurality of target nucleotide sequences present in a nucleic acid or a mixture of nucleic acids, by hybridizations and elongations of a plurality of pairs of amplification primers, wherein the plurality of pairs of amplification primers is a plurality of pairs of sense and antisense composite primers according to claim 9 (see pages 219-220 where Casilli teaches PCR amplification using primers that meet the limitations of the instant claim 9). Specifically, the primers taught by Casilli posses the following characteristics:

(1) a hybridization segment, respectively sense or antisense, which pairs with said nucleic acid or mixture of nucleic acids, so as to constitute a sense or antisense primer for one of the

target nucleotide sequences of the plurality targeted (page 219, "Primer Design" section; see also Table 1 for a listing of the target-specific hybridization segments)

(2) a nucleotide tag which is attached to the 5' end of said hybridization segment, but which does not pair with said nucleic acid or mixture of nucleic acids (page 219, "Primer Design" section; see also legend of Table 1)

(3) optionally, of a non-nucleotide component (page 219, "Primer Design" section teaches that the forward primers contain the FAM fluorophore – a non-nucleotide component).

The primers taught by Casilli also possess the following characteristics:

(4) each sense composite primer has, within said plurality, an antisense composite primer with which it forms a pair of sense and antisense composite primers whose respective hybridization segments constitute, with respect to one another, a pair of sense and antisense primers for one of said target nucleotide sequences, each one of said target nucleotide sequences of the plurality targeted thus having a pair of sense and antisense composite primers which is intended for its amplification (page 219, "Primer Design" section; see also Table 1 on page 220)

(5) all the sense composite primers contain the same nucleotide tag and all the antisense composite primers contain in the same nucleotide, the tag of the sense composite primers being different from that of the antisense composite primers (page 219, "Primer Design" section; see also Table 1 legend on page 220,

(6) the sequence of the tag of the sense composite primers is absent from said nucleic acid or mixture of nucleic acids, or, at the very least, is only present therein at a frequency at least two times less than that predicted statistically for a random sequence of the same length, and the sequence of the tag of the antisense composite primers is absent from said nucleic acid or

mixture of nucleic acids, or, at the very least, is only present therein at a frequency at least two times less than that predicted statistically for a random sequence of the same length (page 219 teaches that the 5' tag sequence added to the forward and reverse primers is extremely rare),

(7) the melting temperature of each composite primer (whether it is a sense or antisense primer) has a value 10 to 15 °C higher than that which its hybridization segment would exhibit when naked without tag (see attached printout "Exhibit A"),

(8) each composite primer of said plurality of pairs has a sequence such that no composite primer of said plurality of pairs can form, with itself or with another composite primer of the same plurality, complete or partial base pairing for which the variation in free energy ΔG ociated with the formation of this possible pairing would be greater than 14 kcal/mol (see attached printout, "Exhibit A")

Further regarding claim 26, the attached printout "Exhibit A" only contains analysis of 4 primers (11aF, 11aR, 2F, and 2R) since this is the minimum required by the claim. Using the Oligo Analyzer 3.0 freely available from Integrated DNA Technologies, the melting temperature of each primer with and without the universal tag was calculated and found to meet the limitations of step d of claim 9. Also, the free energies of hairpin and homodimer formation were calculated for each primer and were found to satisfy the requirements of step e of claim 9. Finally, the free energy of each possible heterodimer was calculated and found to meet the limitations of step e of claim 9. Regarding the use of the Oligo Analyzer software, it is noted that the Primer Premier software, version 5.0 is not freely available. Since the PTO does not have experimental capabilities, the primers were analyzed with a web-based oligonucleotide properties calculator. Since, as discussed in greater detail above, it is not clear how the free

energies were calculated, it is virtually impossible to reproduce the calculations done by Applicant. However, since the primers appear to inherently possess the required thermodynamic characteristics, absent any evidence to the contrary the primers taught by Casilli anticipate the instant claim. MPEP 2112 section V notes, "[T]he PTO can require an applicant to prove that the prior art products do not necessarily or inherently possess the characteristics of his [or her] claimed product. Whether the rejection is based on inherency' under 35 U.S.C. 102, on prima facie obviousness' under 35 U.S.C. 103, jointly or alternatively, the burden of proof is the same...[footnote omitted]." The burden of proof is similar to that required with respect to product-by-process claims. In re Fitzgerald, 619 F.2d 67, 70, 205 USPQ 594, 596 (CCPA 1980) (quoting In re Best, 562 F.2d 1252, 1255, 195 USPQ 430, 433-34 (CCPA 1977))." .

Regarding claim 27, Casilli teaches amplification of 4 targets using the above amplification method (see Table 1 on page 220).

Regarding claim 28, the amplified products generated by the method of Casilli are 434 bp, 297 bp, 336 bp, and 375 bp (see Table 1), thereby anticipating the instant limitation that the amplicon sizes be 90-500 bp.

Regarding claim 29, Casilli teaches inclusion of DMSO in the multiplex amplification reaction (page 220, "PCR conditions" section, especially column 2, which teaches inclusion of DMSO for "multiplex 1").

Regarding claim 30, Casilli teaches that the amplifications of said target nucleotide sequences have exponential phase kinetics (page 220, "PCR conditions" section).

Regarding claim 31, Cassilli teaches that all the pairs of composite primers are used in equimolar concentration (page 220, "PCR conditions" section).

Regarding claim 32, Casilli teaches a method for determining the presence or absence of at least one genomic rearrangement within a genetic material B relative to a reference genetic material A, comprising:

(a) selecting at least one nucleotide target which constitutes a marker for the rearrangement(s) to be detected

(b) applying the amplification method according to claim 26 to genetic material B using for each target selected, a pair of composite primers, wherein material B being considered as exhibiting the genetic rearrangement when the result of amplification of the marker target obtained from the material B, is significantly different from that which is obtained from the reference material A under identical conditions, and wherein material B being considered as not exhibiting the genetic rearrangement when the result of amplification of the marker target obtained from the material B is not significantly different from that which is obtained from the reference material A under identical conditions (see pages 221-223 where Casilli teaches application of the method described above to detection of previously known and novel rearrangements in the human BRCA1 gene. Here, Casilli teaches that rearrangements generate distinguishable amplicons compared to normal controls. See also Figures 1-3).

Regarding claim 33, Casilli teaches that the at least one genomic rearrangement is a gene rearrangement (page 223).

Regarding claim 35, Casilli teaches that genetic material B comprises at least one human gene (page 223, where Casilli analyzes the BRCA1 gene).

Regarding claim 36, Casilli teaches a method for determining at least one of the limits of one or more genomic rearrangement(s) which has (have) been detected within a genetic material

B by comparison with a reference genetic material A (see page 223, where Casilli teaches mapping the 3' and 5' boundaries of deletions), comprising:

(a) selecting a candidate region within which said at least one limit is potentially located (page 223, where Casilli teaches mapping the boundaries of a large heterozygous deletion)

(b) for each rearrangement, choosing a set of nucleotide targets is chosen, among which at least one is chosen to constitute marker for this rearrangement, the other target(s) being chosen on both sides or on one or other sides of this marker target inside the candidate region chosen in step (a) so as to cover the extent of this candidate region (page 223, col. 1, paragraphs 3-4),

(c) applying the method according to claim 26 using for each target of the chosen set, at least one pair of composite primers (page 223, col. 1, paragraphs 3-4),

(d) measuring, for each target, the intensity of amplification thus obtained from the genetic material B and comparing it to the control intensity which is obtained for this same target under the same conditions but by applying the method of amplification to reference genetic material A, (page 223, column 1, paragraphs 3-4; see also Figure 3)

(e) determining whether, within the chosen set of targets, at least one target is amplified with an intensity not significantly different from the control intensity, and, if this is not the case, repeating steps (a) to (e) while broadening the candidate region chosen in step (a), wherein at least one limit of the or of each one of the rearrangements within genetic material B being considered to be within a zone between: the position of the marker target for the rearrangement, and the position of the target which has been amplified with an intensity not significantly different from the control intensity or, if there are several of them, with that which is closest to the marker target, (page 223, paragraphs 3-4; see also Figure 3)

Regarding claim 37, Casilli teaches creating a genomic rearrangement map using the results obtained from the above method (page 223 and Figure 3).

Regarding claim 38, Casilli teaches a method for identifying, and optionally isolating, at least one gene liable to be involved in a genetic disease, comprising:

(a) conducting the method of claim 32 on a genetic material B derived from organisms exhibiting the genetic disease and a genomic material which is comparable but derived from control organisms serving as reference genomic material A, so as to detect the rearrangement(s) present in the material B relative to the material A

(b) the gene(s) affected by the detected rearrangement(s) is (are) identified, and optionally isolated, this (these) identified and optionally isolated gene(s) corresponding to the gene(s) liable to be involved in said genetic disease (see pages 221-223; see also Figures 1 & 2).

11. Claims 26-28, 32, 33, and 35-38 are rejected under 35 U.S.C. 102(b) as being anticipated by Duponchel et al. (Human Mutation (2001) 17: 61-70; cited on IDS).

Regarding claim 26, Duponchel teaches a method for amplifying in multiplex a plurality of target nucleotide sequences present in a nucleic acid or a mixture of nucleic acids, by hybridizations and elongations of a plurality of pairs of amplification primers, wherein the plurality of pairs of amplification primers is a plurality of pairs of sense and antisense composite primers according to claim 9 (see pages 62-63 where Duponchel teaches PCR amplification using primers that meet the limitations of the instant claim 9). Specifically, the primers taught by Duponchel posses the following characteristics:

(1) a hybridization segment, respectively sense or antisense, which pairs with said nucleic acid or mixture of nucleic acids, so as to constitute a sense or antisense primer for one of the target nucleotide sequences of the plurality targeted (page 63, "Indirect Fluorescent Labeling" section; see also Table 1 for a listing of the target-specific hybridization segments)

(2) a nucleotide tag which is attached to the 5' end of said hybridization segment, but which does not pair with said nucleic acid or mixture of nucleic acids (page 63, "Indirect Fluorescent Labeling" section; see also Table 1)

(3) optionally, of a non-nucleotide component (page 63, "Indirect Fluorescent Labeling" section and also Table 1 which teach that the forward primers contain the FAM fluorophore – a non-nucleotide component).

The primers taught by Duponchel also possess the following characteristics:

(4) each sense composite primer has, within said plurality, an antisense composite primer with which it forms a pair of sense and antisense composite primers whose respective hybridization segments constitute, with respect to one another, a pair of sense and antisense primers for one of said target nucleotide sequences, each one of said target nucleotide sequences of the plurality targeted thus having a pair of sense and antisense composite primers which is intended for its amplification (page 63, "Indirect Fluorescent Labeling" section; see also Table 1)

(5) all the sense composite primers contain the same nucleotide tag and all the antisense composite primers contain in the same nucleotide, the tag of the sense composite primers being different from that of the antisense composite primers (page 63, "Indirect Fluorescent Labeling" section; see also Table 1)

(6) the sequence of the tag of the sense composite primers is absent from said nucleic acid or mixture of nucleic acids, or, at the very least, is only present therein at a frequency at least two times less than that predicted statistically for a random sequence of the same length, and the sequence of the tag of the antisense composite primers is absent from said nucleic acid or mixture of nucleic acids, or, at the very least, is only present therein at a frequency at least two times less than that predicted statistically for a random sequence of the same length (page 63, column 1 teaches that the 5' tag sequence added to the forward and reverse primers is extremely rare),

(7) the melting temperature of each composite primer (whether it is a sense or antisense primer) has a value 10 to 15 °C higher than that which its hybridization segment would exhibit when naked without tag (see attached printout "Exhibit B"),

(8) each composite primer of said plurality of pairs has a sequence such that no composite primer of said plurality of pairs can form, with itself or with another composite primer of the same plurality, complete or partial base pairing for which the variation in free energy ΔG ociated with the formation of this possible pairing would be greater than 14 kcal/mol (see attached printout, "Exhibit B").

Further regarding claim 26, the attached printout "Exhibit B" only contains analysis of 4 primers (D8159ex5F, D8578ex5R, BRCA1ex5F, BRCA1ex5R) since this is the minimum required by the claim. Using the Oligo Analyzer 3.0 freely available from Integrated DNA Technologies, the melting temperature of each primer with and without the universal tag was calculated and found to meet the limitations of step d of claim 9. Also, the free energies of hairpin and homodimer formation were calculated for each primer and were found to satisfy the

requirements of step e of claim 9. Finally, the free energy of each possible heterodimer was calculated and found to meet the limitations of step e of claim 9. Regarding the use of the Oligo Analyzer software, it is noted that the Primer Premier software, version 5.0 is not freely available. Since the PTO does not have experimental capabilities, the primers were analyzed with a web-based oligonucleotide properties calculator. Since, as discussed in greater detail above, it is not clear how the free energies were calculated, it is virtually impossible to reproduce the calculations done by Applicant. However, since the primers appear to inherently possess the required thermodynamic characteristics, absent any evidence to the contrary the primers taught by Casilli anticipate the instant claim. MPEP 2112 section V notes, "[T]he PTO can require an applicant to prove that the prior art products do not necessarily or inherently possess the characteristics of his [or her] claimed product. Whether the rejection is based on inherency' under 35 U.S.C. 102, on prima facie obviousness' under 35 U.S.C. 103, jointly or alternatively, the burden of proof is the same...[footnote omitted]." The burden of proof is similar to that required with respect to product-by-process claims. In re Fitzgerald, 619 F.2d 67, 70, 205 USPQ 594, 596 (CCPA 1980) (quoting In re Best, 562 F.2d 1252, 1255, 195 USPQ 430, 433-34 (CCPA 1977))."

Regarding claim 27, Duponchel teaches amplification of 6 targets using the above amplification method (see page 62, column 2 and Table 1).

Regarding claim 28, Duponchel teaches amplified products of 376 bp and 490 bp (see Table 1). Since the claims only require amplification of 2 targets in multiplex, the teachings of Duponchel meet the limitations of claim 28.

Regarding claim 32, Duponchel teaches a method for determining the presence or absence of at least one genomic rearrangement within a genetic material B relative to a reference genetic material A, comprising:

(a) selecting at least one nucleotide target which constitutes a marker for the rearrangement(s) to be detected

(b) applying the amplification method according to claim 26 to genetic material B using for each target selected, a pair of composite primers, wherein material B being considered as exhibiting the genetic rearrangement when the result of amplification of the marker target obtained from the material B, is significantly different from that which is obtained from the reference material A under identical conditions, and wherein material B being considered as not exhibiting the genetic rearrangement when the result of amplification of the marker target obtained from the material B is not significantly different from that which is obtained from the reference material A under identical conditions (see pages 64-68 where Duponchel teaches application of the method described above to detection of previously known and novel rearrangements in the human C1NH gene. Here, Duponchel teaches that rearrangements generate distinguishable amplicons compared to normal controls (pages 67-68; see also Figures 1 & 2)).

Regarding claim 33, Duponchel teaches that the at least one genomic rearrangement is a gene rearrangement (page 64, column 2).

Regarding claim 35, Duponchel teaches that genetic material B comprises at least one human gene (page 64, column 1, where the human C1NH gene was analyzed).

Regarding claim 36, Duponchel teaches a method for determining at least one of the limits of one or more genomic rearrangement(s) which has (have) been detected within a genetic material B by comparison with a reference genetic material A (see page 64, where Duponchel teaches mapping boundaries of the rearrangements occurring in exon 4), comprising:

(a) selecting a candidate region within which said at least one limit is potentially located (page 64, column 1, where Duponchel teaches mapping the boundaries of duplications and deletions in exon 4)

(b) for each rearrangement, choosing a set of nucleotide targets is chosen, among which at least one is chosen to constitute marker for this rearrangement, the other target(s) being chosen on both sides or on one or other sides of this marker target inside the candidate region chosen in step (a) so as to cover the extent of this candidate region (page 64, column 1),

(c) applying the method according to claim 26 using for each target of the chosen set, at least one pair of composite primers (page 64, column 1),

(d) measuring, for each target, the intensity of amplification thus obtained from the genetic material B and comparing it to the control intensity which is obtained for this same target under the same conditions but by applying the method of amplification to reference genetic material A, (page 64, column 1)

(e) determining whether, within the chosen set of targets, at least one target is amplified with an intensity not significantly different from the control intensity, and, if this is not the case, repeating steps (a) to (e) while broadening the candidate region chosen in step (a), wherein at least one limit of the or of each one of the rearrangements within genetic material B being considered to be within a zone between: the position of the marker target for the rearrangement,

and the position of the target which has been amplified with an intensity not significantly different from the control intensity or, if there are several of them, with that which is closest to the marker target, (page 64)

Regarding claim 37, Duponchel teaches creating a genomic rearrangement map using the results obtained from the above method (page 64, column 1 – column 2).

Regarding claim 38, Duponchel teaches a method for identifying, and optionally isolating, at least one gene liable to be involved in a genetic disease, comprising:

(a) conducting the method of claim 32 on a genetic material B derived from organisms exhibiting the genetic disease and a genomic material which is comparable but derived from control organisms serving as reference genomic material A, so as to detect the rearrangement(s) present in the material B relative to the material A

(b) the gene(s) affected by the detected rearrangement(s) is (are) identified, and optionally isolated, this (these) identified and optionally isolated gene(s) corresponding to the gene(s) liable to be involved in said genetic disease (see 64-68 and also Figures 1-2).

Claim Rejections - 35 USC § 103

12. The following is a quotation of 35 U.S.C. 103(a) which forms the basis for all obviousness rejections set forth in this Office action:

> (a) A patent may not be obtained though the invention is not identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains. Patentability shall not be negatived by the manner in which the invention was made.

This application currently names joint inventors. In considering patentability of the claims under 35 U.S.C. 103(a), the examiner presumes that the subject matter of the various claims was commonly owned at the time any inventions covered therein were made absent any evidence to the contrary. Applicant is advised of the obligation under 37 CFR 1.56 to point out the inventor and invention dates of each claim that was not commonly owned at the time a later invention was made in order for the examiner to consider the applicability of 35 U.S.C. 103(c) and potential 35 U.S.C. 102(e), (f) or (g) prior art under 35 U.S.C. 103(a).

13. Claims 29-31 are rejected under 35 U.S.C. 103(a) as being unpatentable over Duponchel et al. (Human Mutation (2001) 17: 61-70; cited on IDS) in view of Varadaraj et al. (Gene (1994) 140: 1-5).

Duponchel teaches the method of claim 26, as discussed above.

Regarding claim 30, Duponchel teaches that the amplifications of said target nucleotide sequences have exponential phase kinetics (page 63-64).

Regarding claim 31, Duponchel teaches that six of the twelve primers included in the multiplex PCR reaction mixture were used at equimolar concentrations, whereas the other six primers were used at different concentrations (see page 62, column 2).

Duponchel does not teach inclusion of an agent that facilitates strand separation, such as DMSO, in the amplification reaction, nor does Duponchel teach that all primers were used at equimolar concentrations.

Varadaraj teaches methods of improving PCR amplification of GC-rich templates (see abstract). Regarding claim 29, Varadaraj teaches that DMSO is known to improve DNA sequencing reactions by reducing inter- and intra-strand reannealing (page 3). Varadaraj also cites two instances where the inclusion of DMSO was required to obtain a detectable amplification product (pages 3-4). Varadaraj further teaches that inclusion of DMSO may improve the specificity of PCR reactions (page 4). Finally, Varadaraj concludes, "With the SF, glycerol, DMSO, and NP-40 produced the most specific product and resulted in the greatest amplification of the segment of the G+C-rich satellite DNA [sic] (page 4)."

It would have been prima facie obvious for one of ordinary skill in the art at the time of invention to include DMSO in the multiplex amplification method taught by Duponchel. Varadaraj expressly taught that inclusion of DMSO in PCR reactions improved the specificity of the reaction and also increased the yield by reducing inter- and intra-strand reannealing (pages 3-4). An ordinary practitioner would have been motivated by these teachings of Varadaraj to include DMSO in the PCR reaction mixture in order to improve the product yield and specificity of the reaction. An ordinary practitioner would have expected a reasonable level of success in adding DMSO to the reaction mixture taught by Duponchel, because Varadaraj expressly taught useful concentrations of DMSO to be used with the AmpliTaq enzyme used in the Duponchel method (page 4, column 1). Therefore, an ordinary practitioner of the multiplex amplification method taught by Duponchel, interested in increasing the yield by reducing strand reannealing and also improving the specificity of the reaction, would have been motivated to include DMSO

in the reaction mixture, as suggested by Varadaraj, thus resulting in the instantly claimed method.

It would also have been prima facie obvious for one of ordinary skill in the art at the time of invention to use all of the primers at equimolar concentrations. As noted in *In re Aller*, 105 USPQ 233 at 235:

> More particularly, where the general conditions of a claim are disclosed in the prior art, it is not inventive to discover the optimum or workable ranges by routine experimentation.

Routine optimization is not considered inventive and no evidence has been presented that the selection of the claimed primer concentrations was other than routine or that the results should be considered unexpected in any way as compared to the closest prior art. Therefore, the method of claim 31 is prima facie obvious in view of the cited references.

14. Claim 34 is rejected under 35 U.S.C. 103(a) as being unpatentable over Duponchel et al. (Human Mutation (2001) 17: 61-70; cited on IDS) in view of Meier et al. (American Journal of Pathology (2001) 159(6): 2031-2043).

Duponchel teaches the method of claim 32, as discussed above.

Duponchel does not teach application of the method to chromosomal rearrangement.

Meier teaches a method for detecting chromosomal rearrangements comprising multiplex PCR and capillary electrophoresis (see abstract and page 2034). Meier teaches that "molecular diagnosis of B- and T-cell non-Hodgkin lymphomas are based on determination of clonality of the respective antigen receptors and detection of specific chromosomal translocations (page

2031, column 2)." Meier further teaches that multiplex PCR assays have made detection of these translocations faster and more efficient (page 2032, column 1; see also page 2033, column 2).

It would have been prima facie obvious for one of ordinary skill in the art at the time of invention to apply the method taught by Duponchel to the detection of chromosomal rearrangements. Meier expressly taught that detection of chromosomal rearrangements was an important molecular diagnostic criterion for B- and T-cell non-Hodgkin lymphomas (page 2031). Since Meier expressly advocated detecting chromosomal rearrangements using multiplex PCR (pages 2032-2033), an ordinary practitioner would have been motivated to apply the method taught by Duponchel to the detection of chromosomal rearrangements in order to further expand the applicability of the method with a reasonable expectation of success.

Conclusion

No claims are currently allowable.

Any inquiry concerning this communication or earlier communications from the examiner should be directed to Angela Bertagna whose telephone number is 571-272-8291. The examiner can normally be reached on M-F, 7:30 - 5.

If attempts to reach the examiner by telephone are unsuccessful, the examiner's supervisor, Gary Benzion can be reached on 571-272-0782. The fax phone number for the organization where this application or proceeding is assigned is 571-273-8300.

Information regarding the status of an application may be obtained from the Patent Application Information Retrieval (PAIR) system. Status information for published applications may be obtained from either Private PAIR or Public PAIR. Status information for unpublished applications is available through Private PAIR only. For more information about the PAIR system, see http://pair-direct.uspto.gov. Should you have questions on access to the Private PAIR system, contact the Electronic Business Center (EBC) at 866-217-9197 (toll-free). If you would like assistance from a USPTO Customer Service Representative or access to the automated information system, call 800-786-9199 (IN USA OR CANADA) or 571-272-1000.

JEFFREY FREDMAN
PRIMARY EXAMINER
11/9/06

Angela Bertagna
Examiner, Art Unit 1637
November 8, 2006
amb

Attachments: Exhibit A and Exhibit B

[Exhibit A]

Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- ACC GTT AGT AGT CGA CTG ATT TGA ACA CCA CTG AGA -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na^+ Conc: 50 mM

ANALYZE | HAIRPIN | SELF-DIMER | HETERO-DIMER | NCBI BLAST | TM MISMATCH | LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

DILUTION | RESUSPENSION

RESULTS

SEQUENCE:

5'- ACC GTT AGT AGT CGA CTG ATT TGA ACA CCA CTG AGA -3'

COMPLEMENT:

5'- TCT CAG TGG TGT TCA AAT CAG TCG ACT ACT AAC GGT -3'

LENGTH:	36
GC CONTENT:	44.4 %
MELT TEMP:	63.6 °C
MOLECULAR WEIGHT:	11068.2 g/mole
EXTINCTION COEFFICIENT:	357500 L/(mole·cm)
nmole/OD_{260}:	2.80
µg/OD_{260}:	30.96

MELTING TEMPERATURE SETTINGS

TARGET TYPE:	DNA
OLIGO CONC	0.25 µM
Na^+ CONC	50 mM monovalent salt

MELTING TEMPERATURE ASSUMPTIONS AND LIMITATIONS

- Predictions are accurate for oligos from 8 to 60 bases in length, in neutral buffered solutions (pH 7 - 8) with monovalent cation concentrations from 10mM to 1.2 M.
- Oligo concentration is assumed to be significantly larger (at least 6x) than concentration of the complementary target, which is true in majority of molecular biology experiments. If this is not a case, concentration of the target cannot be ignored and you should enter in the box,

  Oligo Conc = [strand1] - [strand2]/2 when [strand1] ⩾ [strand2]
  Oligo Conc = ([strand1] + [strand2])/4 when [strand1] = [strand2]

- Melting temperature accuracy and models: (Oligo/Template)

  DNA/DNA: +/- 1.4°C (Allawi '97)
  LNA/DNA: +/- 2.0°C (McTigue '04)
  RNA/DNA: +/- 2.7°C (Sugimoto '95)
  DNA/RNA: +/- 2.7°C (Sugimoto '95)
  RNA/RNA: +/- 1.3°C (Xia '98)

Monovalent cation correction: +/- 2.0°C (Owczarzy '04)

- Non-consecutive LNA bases hybridized to a DNA template use a model from McTigue '04. Consecutive LNA bases on a DNA template and any LNA bases on an RNA template assume RNA energetic parameters and predictions are therefore less accurate.
- Effects of chemical modifications are neglected except when the modification contains a base, e.g., 5-Methyl dC, Internal Fluorescein dT. Energetic effects of these modifications are only approximated.
- This OligoAnalyzer version does not account for effects of divalent cations.

Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- TGA TTT GAA CAC CAC TGA GA -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na^+ Conc: 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

DILUTION | RESUSPENSION

RESULTS

SEQUENCE:
5'- TGA TTT GAA CAC CAC TGA GA -3'
COMPLEMENT:
5'- TCT CAG TGG TGT TCA AAT CA -3'

LENGTH:	20
GC CONTENT:	40.0 %
MELT TEMP:	51.5 °C
MOLECULAR WEIGHT:	6125.0 g/mole
EXTINCTION COEFFICIENT:	199600 L/(mole·cm)
nmole/OD_{260}:	5.01
µg/OD_{260}:	30.69

MELTING TEMPERATURE SETTINGS

TARGET TYPE:	DNA
OLIGO CONC	0.25 µM
Na^+ CONC	50 mM monovalent salt

MELTING TEMPERATURE ASSUMPTIONS AND LIMITATIONS

- Predictions are accurate for oligos from 8 to 60 bases in length, in neutral buffered solutions (pH 7 - 8) with monovalent cation concentrations from 10mM to 1.2 M.
- Oligo concentration is assumed to be significantly larger (at least 6x) than concentration of the complementary target, which is true in majority of molecular biology experiments. If this is not a case, concentration of the target cannot be ignored and you should enter in the box,

  Oligo Conc = [strand1] - [strand2]/2 when [strand1] ⩾ [strand2]
  Oligo Conc = ([strand1] + [strand2])/4 when [strand1] = [strand2]

- Melting temperature accuracy and models: (Oligo/Template)

  DNA/DNA: +/- 1.4°C (Allawi '97)
  LNA/DNA: +/- 2.0°C (McTigue '04)
  RNA/DNA: +/- 2.7°C (Sugimoto '95)
  DNA/RNA: +/- 2.7°C (Sugimoto '95)
  RNA/RNA: +/- 1.3°C (Xia '98)

Monovalent cation correction: +/- 2.0°C (Owczarzy '04)

- Non-consecutive LNA bases hybridized to a DNA template use a model from McTigue '04. Consecutive LNA bases on a DNA template and any LNA bases on an RNA template assume RNA energetic parameters and predictions are therefore less accurate.
- Effects of chemical modifications are neglected except when the modification contains a base, e.g., 5-Methyl dC, Internal Fluorescein dT. Energetic effects of these modifications are only approximated.
- This OligoAnalyzer version does not account for effects of divalent cations.

Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- TCG GAT AGC TAG TCG TCC GCC TAT CAT TAC ATG TTT -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na+ Conc: 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

DILUTION | RESUSPENSION

RESULTS

SEQUENCE:

5'- TCG GAT AGC TAG TCG TCC GCC TAT CAT TAC ATG TTT -3'

COMPLEMENT:

5'- AAA CAT GTA ATG ATA GGC GGA CGA CTA GCT ATC CGA -3'

LENGTH:	36
GC CONTENT:	44.4 %
MELT TEMP:	63.5 °C
MOLECULAR WEIGHT:	10992.2 g/mole
EXTINCTION COEFFICIENT:	337600 L/(mole·cm)
nmole/OD_{260}:	2.96
µg/OD_{260}:	32.56

MELTING TEMPERATURE SETTINGS

TARGET TYPE:	DNA
OLIGO CONC	0.25 µM
Na^+ CONC	50 mM monovalent salt

MELTING TEMPERATURE ASSUMPTIONS AND LIMITATIONS

- Predictions are accurate for oligos from 8 to 60 bases in length, in neutral buffered solutions (pH 7 - 8) with monovalent cation concentrations from 10mM to 1.2 M.
- Oligo concentration is assumed to be significantly larger (at least 6x) than concentration of the complementary target, which is true in majority of molecular biology experiments. If this is not a case, concentration of the target cannot be ignored and you should enter in the box,

  Oligo Conc = [strand1] - [strand2]/2 when [strand1] ⩾ [strand2]
  Oligo Conc = ([strand1] + [strand2])/4 when [strand1] = [strand2]

- Melting temperature accuracy and models: (Oligo/Template)

  DNA/DNA: +/- 1.4°C (Allawi '97)
  LNA/DNA: +/- 2.0°C (McTigue '04)
  RNA/DNA: +/- 2.7°C (Sugimoto '95)
  DNA/RNA: +/- 2.7°C (Sugimoto '95)
  RNA/RNA: +/- 1.3°C (Xia '98)

Monovalent cation correction: +/- 2.0°C (Owczarzy '04)

- Non-consecutive LNA bases hybridized to a DNA template use a model from McTigue '04. Consecutive LNA bases on a DNA template and any LNA bases on an RNA template assume RNA energetic parameters and predictions are therefore less accurate.
- Effects of chemical modifications are neglected except when the modification contains a base, e.g., 5-Methyl dC, Internal Fluorescein dT. Energetic effects of these modifications are only approximated.
- This OligoAnalyzer version does not account for effects of divalent cations.

Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- CCG CCT ATC ATT ACA TGT TT -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na+ Conc: 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

DILUTION | RESUSPENSION

RESULTS

SEQUENCE:
5'- CCG CCT ATC ATT ACA TGT TT -3'

COMPLEMENT:
5'- AAA CAT GTA ATG ATA GGC GG -3'

LENGTH:	20
GC CONTENT:	40.0 %
MELT TEMP:	50.3 °C
MOLECULAR WEIGHT:	6018.0 g/mole
EXTINCTION COEFFICIENT:	181900 L/(mole·cm)
nmole/OD_{260}:	5.50
µg/OD_{260}:	33.08

MELTING TEMPERATURE SETTINGS

TARGET TYPE:	DNA
OLIGO CONC	0.25 µM
Na^+ CONC	50 mM monovalent salt

MELTING TEMPERATURE ASSUMPTIONS AND LIMITATIONS

- Predictions are accurate for oligos from 8 to 60 bases in length, in neutral buffered solutions (pH 7 - 8) with monovalent cation concentrations from 10mM to 1.2 M.
- Oligo concentration is assumed to be significantly larger (at least 6x) than concentration of the complementary target, which is true in majority of molecular biology experiments. If this is not a case, concentration of the target cannot be ignored and you should enter in the box,

  Oligo Conc = [strand1] - [strand2]/2 when [strand1] ≥ [strand2]
  Oligo Conc = ([strand1] + [strand2])/4 when [strand1] = [strand2]

- Melting temperature accuracy and models: (Oligo/Template)

  DNA/DNA: +/- 1.4°C (Allawi '97)
  LNA/DNA: +/- 2.0°C (McTigue '04)
  RNA/DNA: +/- 2.7°C (Sugimoto '95)
  DNA/RNA: +/- 2.7°C (Sugimoto '95)
  RNA/RNA: +/- 1.3°C (Xia '98)

Monovalent cation correction: +/- 2.0°C (Owczarzy '04)

- Non-consecutive LNA bases hybridized to a DNA template use a model from McTigue '04. Consecutive LNA bases on a DNA template and any LNA bases on an RNA template assume RNA energetic parameters and predictions are therefore less accurate.
- Effects of chemical modifications are neglected except when the modification contains a base, e.g., 5-Methyl dC, Internal Fluorescein dT. Energetic effects of these modifications are only approximated.
- This OligoAnalyzer version does not account for effects of divalent cations.

Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- ACC GTT AGT AGT CGA CGG ACG TTG TCA TTA GTT CTT TGG -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na+ Conc: 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

DILUTION | RESUSPENSION

RESULTS

SEQUENCE:
5'- ACC GTT AGT AGT CGA CGG ACG TTG TCA TTA GTT CTT TGG -3'

COMPLEMENT:
5'- CCA AAG AAC TAA TGA CAA CGT CCG TCG ACT ACT AAC GGT -3'

LENGTH:	39
GC CONTENT:	46.2 %
MELT TEMP:	64.7 °C
MOLECULAR WEIGHT:	12034.8 g/mole
EXTINCTION COEFFICIENT:	374600 L/(mole·cm)
nmole/OD_{260}:	2.67
µg/OD_{260}:	32.13

MELTING TEMPERATURE SETTINGS

TARGET TYPE:	DNA
OLIGO CONC	0.25 µM
Na^+ CONC	50 mM monovalent salt

MELTING TEMPERATURE ASSUMPTIONS AND LIMITATIONS

- Predictions are accurate for oligos from 8 to 60 bases in length, in neutral buffered solutions (pH 7 - 8) with monovalent cation concentrations from 10mM to 1.2 M.
- Oligo concentration is assumed to be significantly larger (at least 6x) than concentration of the complementary target, which is true in majority of molecular biology experiments. If this is not a case, concentration of the target cannot be ignored and you should enter in the box,

  Oligo Conc = [strand1] - [strand2]/2 when [strand1] ⩾ [strand2]
  Oligo Conc = ([strand1] + [strand2])/4 when [strand1] = [strand2]

- Melting temperature accuracy and models: (Oligo/Template)

  DNA/DNA: +/- 1.4°C (Allawi '97)
  LNA/DNA: +/- 2.0°C (McTigue '04)
  RNA/DNA: +/- 2.7°C (Sugimoto '95)
  DNA/RNA: +/- 2.7°C (Sugimoto '95)
  RNA/RNA: +/- 1.3°C (Xia '98)

Monovalent cation correction: +/- 2.0°C (Owczarzy '04)

- Non-consecutive LNA bases hybridized to a DNA template use a model from McTigue '04. Consecutive LNA bases on a DNA template and any LNA bases on an RNA template assume RNA energetic parameters and predictions are therefore less accurate.
- Effects of chemical modifications are neglected except when the modification contains a base, e.g., 5-Methyl dC, Internal Fluorescein dT. Energetic effects of these modifications are only approximated.
- This OligoAnalyzer version does not account for effects of divalent cations.

Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- GGA CGT TGT CAT TAG TTC TTT GG -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na^+ Conc: 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

DILUTION | RESUSPENSION

RESULTS

SEQUENCE:
5'- GGA CGT TGT CAT TAG TTC TTT GG -3'
COMPLEMENT:
5'- CCA AAG AAC TAA TGA CAA CGT CC -3'

LENGTH:	23
GC CONTENT:	43.5 %
MELT TEMP:	54.0 °C
MOLECULAR WEIGHT:	7091.6 g/mole
EXTINCTION COEFFICIENT:	216700 L/(mole·cm)
nmole/OD_{260}:	4.61
µg/OD_{260}:	32.73

MELTING TEMPERATURE SETTINGS

TARGET TYPE:	DNA
OLIGO CONC	0.25 µM
Na^+ CONC	50 mM monovalent salt

MELTING TEMPERATURE ASSUMPTIONS AND LIMITATIONS

- Predictions are accurate for oligos from 8 to 60 bases in length, in neutral buffered solutions (pH 7 - 8) with monovalent cation concentrations from 10mM to 1.2 M.
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  LNA/DNA: +/- 2.0°C (McTigue '04)
  RNA/DNA: +/- 2.7°C (Sugimoto '95)
  DNA/RNA: +/- 2.7°C (Sugimoto '95)
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Monovalent cation correction: +/- 2.0°C (Owczarzy '04)

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Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- TCG GAT AGC TAG TCG TAC ATG TCT TTT CTT CCC TAG TAT G -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na+ Conc: 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

DILUTION | RESUSPENSION

RESULTS

SEQUENCE:

5'- TCG GAT AGC TAG TCG TAC ATG TCT TTT CTT CCC TAG TAT G -3'

COMPLEMENT:

5'- CAT ACT AGG GAA GAA AAG ACA TGT ACG ACT AGC TAT CCG A -3'

LENGTH:	40
GC CONTENT:	42.5 %
MELT TEMP:	62.6 °C
MOLECULAR WEIGHT:	12234.0 g/mole
EXTINCTION COEFFICIENT:	374000 L/(mole·cm)
nmole/OD_{260}:	2.67
µg/OD_{260}:	32.71

MELTING TEMPERATURE SETTINGS

TARGET TYPE:	DNA
OLIGO CONC	0.25 µM
Na^+ CONC	50 mM monovalent salt

MELTING TEMPERATURE ASSUMPTIONS AND LIMITATIONS

- Predictions are accurate for oligos from 8 to 60 bases in length, in neutral buffered solutions (pH 7 - 8) with monovalent cation concentrations from 10mM to 1.2 M.
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Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- ACA TGT CTT TTC TTC CCT AGT ATG -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na^+ Conc: 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

DILUTION | RESUSPENSION

RESULTS

SEQUENCE:
5'- ACA TGT CTT TTC TTC CCT AGT ATG -3'
COMPLEMENT:
5'- CAT ACT AGG GAA GAA AAG ACA TGT -3'

LENGTH:	24
GC CONTENT:	37.5 %
MELT TEMP:	52.3 °C
MOLECULAR WEIGHT:	7259.8 g/mole
EXTINCTION COEFFICIENT:	219100 L/(mole·cm)
nmole/OD_{260}:	4.56
µg/OD_{260}:	33.13

MELTING TEMPERATURE SETTINGS

TARGET TYPE:	DNA
OLIGO CONC	0.25 µM
Na^+ CONC	50 mM monovalent salt

MELTING TEMPERATURE ASSUMPTIONS AND LIMITATIONS

- Predictions are accurate for oligos from 8 to 60 bases in length, in neutral buffered solutions (pH 7 - 8) with monovalent cation concentrations from 10mM to 1.2 M.
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  RNA/DNA: +/- 2.7°C (Sugimoto '95)
  DNA/RNA: +/- 2.7°C (Sugimoto '95)
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Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- ACC GTT AGT AGT CGA CGG ACG TTG TCA TTA GTT CTT TGG -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na^+ Conc: 50 mM

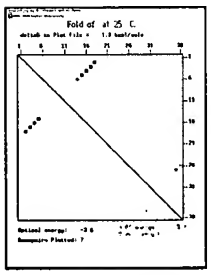
ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

mFold Input

General Information

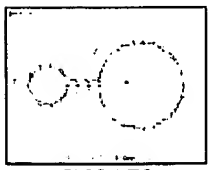


CLICK TO VIEW DOT PLOT

Sequence Name: 39 Base Oligo
Batch Date: 11/8/2006
Sequence :
ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG

ADD TO ORDER

Structure 1



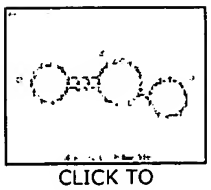
CLICK TO ENLARGE

ΔG -3.69 kcal.mole^{-1}
T_M 56.2 °C
ΔH -39.1 kcal.mole^{-1}
ΔS -118.7 cal.K^{-1}mole^{-1}

CONNECTIVITY | DETAILS

Structure 2

ΔG -2.39 kcal.mole^{-1}



CLICK TO ENLARGE

T_M	37.9	°C
ΔH	-57.9	kcal.mole^{-1}
ΔS	-186.1	cal.K^{-1}mole^{-1}

CONNECTIVITY DETAILS

Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- ACC GTT AGT AGT CGA CGG ACG TTG TCA TTA GTT CTT TGG -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na+ Conc: 50 mM

ANALYZE | HAIRPIN | SELF-DIMER | HETERO-DIMER | NCBI BLAST | TM MISMATCH | LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

HOMO-DIMER ANALYSIS

Dimer Sequence
5'- ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG -3'

Maximum Delta G -71.89 kcal/mole

Delta G -9.45 kcal/mole
Base Pairs 6

```
5'                ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                    :        ||||||        :
3' GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -8.02 kcal/mole
Base Pairs 4

```
5'                          ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                            ||||          ::::
3' GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -6.3 kcal/mole
Base Pairs 4

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
    ::                ||||              ::
3'  GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -4.95 kcal/mole
Base Pairs 3

```
5'                    ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                      : |||  :: : : ::  ::: :
3' GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -4.95 kcal/mole
Base Pairs 3

```
5'    ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
      :          : :.|||  ::: : :          :
3' GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                                    ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                      : || :
3' GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                           ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                               ||         ::
3' GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'            ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
              :  :  :     || ::     :  :  :
3' GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'          ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
            ::   :   :     ||     :   :   ::
3' GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'        ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
          : :    :     || : : ::    :     : :
3' GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -2.92 kcal/mole
Base Pairs 3

```
5'           ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                       |||     :::
3' GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -2.92 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
    :    :     |||        :::       :    :
3' GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -2.92 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
         :     : :  |||   :::  : :     :
3'     GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                 :       || ::       :
3'            GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                              ||         ::
3'                           GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
     :   || : :       :   :       : : ::   :
3'   GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
           ||                        ::
3'       GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                           :   ||  ::   :
3'                         GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                ||   :          :   ::
3'          GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
               : :  ||  :  : :  :  ::  : :
3'             GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                  :  : :   : ||  :   : :  :
3'                GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                                    ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                      || : :
3' GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                                ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                  ||      : :
3' GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                             ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                               ||    : :   : :
3' GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'              ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                     ||          : :
3' GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                    ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                      ||                    : :
3' GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                  ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                    ||                    : :
3' GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
            :    ||  : :    : :  ::     :
3'         GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                : : : ||           :: : : :
3'             GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'                            ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                               :    || ::    :
3' GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'                          ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                             :     : || :     :
3' GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'      ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
        :     || :                  : ::     :
3' GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'  ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
          : || :                : :: :
3' GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                        :       : || :       :
```

3' GGTTTCTTGATTACTGTTGCAGGCAGCTGATGATTGCCA

For questions regarding the Dimer Analysis contact our Technical Support Group
1-800-328-2661 or e-mail TechSupport@idtdna.com

Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- ACC GTT AGT AGT CGA CGG ACG TTG TCA TTA GTT CTT TGG -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na+ Conc: 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

Primary Sequence
5'- ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG -3'

Secondary Sequence
5'- ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA -3'

Maximum Delta G -71.89 kcal/mole

Delta G -11.04 kcal/mole
Base Pairs 7

```
5'              ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                :  ||||||         :
3'  AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -8.02 kcal/mole
Base Pairs 4

```
5'                      ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                        : ::          ||||
3'  AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -5.02 kcal/mole
Base Pairs 3

```
5'  ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
```

```
                                    : :  |||
3'                                  AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -4.95 kcal/mole
Base Pairs 3

```
5'                ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                      : ::: : : ::  ||| :
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -4.86 kcal/mole
Base Pairs 4

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                            :       ||||
3'                        AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                                  ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                    : || :
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                        ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                            ||          : :
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'          ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
              : : :   :   : ||     :  :  :
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'     ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
              ::       : : ||     :     : :
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -3.53 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                      :       ||| : :      :
3'         AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -3.53 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                    : :   ::   |||
3'              AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -3.53 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                           : ||| : ::   :
3'                         AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -3.3 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                         : |||     :     :
3'                AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -3.29 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                    :   :  |||   :: :::     :
3'             AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -3.17 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                    |||  :
3'                                  AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
       :              :  :                ||
3'     AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
              ::   ||   :              :
3'            AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -2.94 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
       : ||| ::   :  :        : :: :
3'   AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -2.94 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
        :   |||:         :       : : ::   :
3'      AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -2.94 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                  : |||  : :
3'                              AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -2.92 kcal/mole

Base Pairs 3

```
5'        ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
              :   : ::     |||
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -2.92 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
     :   :  :    :    |||      :    :
3'    AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                :       ||
3'                              AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                    :  ||   ::
3'                AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                  : : ||
3'                             AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                ::      ||
3'                              AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
           :    :  :                  ||
3'          AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                       :: ::  : :  ||  : :
3'                   AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'  ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
      :  : ::  || :    :        : ::     :
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
              ||   :     :   : :   ::
3'            AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                  :                    ||
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'              ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                  :                    ||
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'      ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
   .    :  : :  :        :      :   :   ||
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                                  ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                    ||  ::
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                                ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                  ||      ::
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'           ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                  : :          ||
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                          ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                            ||   ::   ::
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
   :     :  ||  :     :: : :          :
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'       ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
           ||     :     :       :  : :
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'   ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
      : ||          :          :  : :
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                       :   ||  :       :  :
3'                     AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'                        ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                          :   || : :   :
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'                      ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                           : || :     :
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

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Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- ACC GTT AGT AGT CGA CGG ACG TTG TCA TTA GTT CTT TGG -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na+ Conc: 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

Primary Sequence
5'- ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG -3'

Secondary Sequence
5'- TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG -3'

Maximum Delta G -71.89 kcal/mole

Delta G -6.68 kcal/mole
Base Pairs 3

```
5'                                    ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                      |||
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -6.53 kcal/mole
Base Pairs 4

```
5'                ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                  :    :  ||||.  :  :   :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -5.19 kcal/mole
Base Pairs 3

```
5'                                ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
```

```
                                      :   ::  |||
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -4.95 kcal/mole
Base Pairs 3

```
5'             ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
               :      ::    :  |||             :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -4.95 kcal/mole
Base Pairs 3

```
5'         ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
           :  :      :    : : |||       :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -4.64 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
             :  : : ||| :  '  :   :: :
3'           GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                             ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                       :   ||
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                         ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                           ::   ::  : : :     ||
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                              ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                  ||
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -3.43 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
           :: : :          : |||        : :
3'        GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -3.43 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                               |||          :
3'                             GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -3.3 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
    :          :  ::         |||          :: :
3'  GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                   : ::  ||
3'                                 GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
              :    ||           :  :
3'           GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                    :  : ||
3'                                  GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -2.92 kcal/mole
Base Pairs 3

```
5'           ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
               :  :  :   :  |||    :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -2.92 kcal/mole
Base Pairs 3

```
5'      ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
          :  :  :   |||          :    : :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -2.92 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
       :::       :  |||  :   :          :  :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5'                          ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                            ::   |||   ::
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5'                        ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                              : |||    : ::
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole

Base Pairs 3

```
5'  ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
    :   :  : :    : :      :   : ||| : :  :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
       :: |||   :   : :   :        :   :
3'    GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                              : |||
3'                          GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -2.3 kcal/mole
Base Pairs 3

```
5'                ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                  :      |||       :       :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                  :  ::   :  :  :      ||
3'                GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
         :      :   ::          ::   ||
3'       GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5'          ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                  :  ||  :          :   :   :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                   :      :      :  ||    : :
3'                 GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5'        ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
            :  ||  :         :        :    : :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5'    ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
        :     : ||    :    : : :       :    :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                   ||
3'                      GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'               ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                 :          :           : : ||
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'     ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
       : :                        : :       ||
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                || : :          :   :
3'      GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5'           ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                   :                : :  ||   :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                                ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                  ||       :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                        ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                             ||  :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                        :   ||       :  :   :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                ||     :  :  :  :  :  :  :
3'              GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'                                  ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                       : ||    :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                    || :       :
3'                                GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

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Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- ACC GTT AGT AGT CGA CGG ACG TTG TCA TTA GTT CTT TGG -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na^+ Conc: 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

Primary Sequence
5'- ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG -3'

Secondary Sequence
5'- TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT -3'

Maximum Delta G -71.89 kcal/mole

Delta G -13.21 kcal/mole
Base Pairs 6

```
5'   ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
     ::   :: :  : : ||||||      :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -6.68 kcal/mole
Base Pairs 3

```
5'                                  ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                    |||
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -6.53 kcal/mole
Base Pairs 4

```
5'            ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
```

```
            :   :   :  ||||   :   :   :
3'  TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -6.53 kcal/mole
Base Pairs 4

```
5'        ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
              :  :   :  ||||            :
3'  TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -5.19 kcal/mole
Base Pairs 3

```
5'                          ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                               :   ::  |||
3'  TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                      ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                 :   ||
3'  TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                  ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                    :      :  : : :      ||
3'  TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                          ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                             ||
3'  TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                  ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                      ||    :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'       ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
           :  ::: ||   :::     :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
      :       ::  : ||       :    :
3'   TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.43 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                           |||    :  : ::
3'                       TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.3 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                     :   ||| :: :
3'                  TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                           :     : :    ||
3'                      TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
          ::: :     ||  :  : :      ::
3'  TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5'                        ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                          ::   |||   ::
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5'                      ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                           : |||    : ::
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
           :                   : ||| : :  :
3'     TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5'          ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
            :  : |||  :       :  :   :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                          : .   |||     :
3'                  TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.95 kcal/mole

Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                  : :      ||  :   :  :
3'              TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                :   :    ||
3'                              TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                          ||
3'                                    TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
         :  :    : :   :  :          || :
3'       TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
            :  :     : : :      :   ||
3'            TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'          ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                                  : : ||
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
          :    :  :  ||        :  : :   :
3'         TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5'      ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
        :       :               : :  ||  :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                  :   : ::  : ||  :
3'                TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                           ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                             ||       :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'     ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
         || :  :   :  :      :  :  :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
         :   ||       : :  : :          :  :
3'      TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                              :   ||  :
3'                           TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                     ||  :           :    :
3'                  TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'                           ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                               : ||   :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'  ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
    : : :     :       :       :   ||
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACGGACGTTGTCATTAGTTCTTTGG
                           .      ||  :  :  :
3'                        TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

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Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- ACC GTT AGT AGT CGA CTG ATT TGA ACA CCA CTG AGA -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na^+ Conc: 50 mM

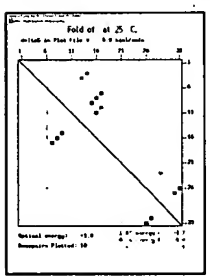
ANALYZE | HAIRPIN | SELF-DIMER | HETERO-DIMER | NCBI BLAST | TM MISMATCH | LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

mFold Input

General Information

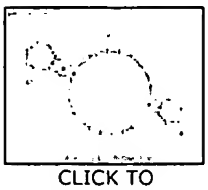


CLICK TO VIEW DOT PLOT

Sequence Name: 36 Base Oligo
Batch Date: 11/8/2006
Sequence :
ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA

ADD TO ORDER

Structure 1



CLICK TO ENLARGE

ΔG	-1.06	kcal.mole^{-1}
T_M	33.0	°C
ΔH	-42.3	kcal.mole^{-1}
ΔS	-138.2	cal.K^{-1}mole^{-1}

CONNECTIVITY | DETAILS

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OligoAnalyzer 3.0

Bases

5'- ACC GTT AGT AGT CGA CTG ATT TGA ACA CCA CTG AGA -3'

Target Type: DNA
Oligo Conc: 0.25 µM
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ANALYZE | HAIRPIN | SELF-DIMER | HETERO-DIMER | NCBI BLAST | TM MISMATCH | LNA CONVERSION

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RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

HOMO-DIMER ANALYSIS

Dimer Sequence
5'- ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA -3'

Maximum Delta G -62.48 kcal/mole

Delta G -12.64 kcal/mole
Base Pairs 8

```
5'              ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                    :  ||||||||  :
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                                  ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                                    :  ||  :
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                          ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                              ||          : :
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -3.29 kcal/mole
Base Pairs 3

```
5'          ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
               |||    :    : :    :    :::
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -2.94 kcal/mole
Base Pairs 3

```
5'                 ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                      : ||| : : ::: :
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -2.94 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
       : ||| ::    :  ::  :    :: ::: :
3'    AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -2.94 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
        :   ||| .:  :        :  : :::   :
3'       AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                    || :    : ::
3'          AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                   : :  ||  ::    : :
3'               AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                : || : :  : : :: :
3'           AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                        ||      ::
3'        .          AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                            ||     ::
3'                        AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                                ||  ::
3'                             AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
           :    :      ||  ::      :    :
3'          AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
              ::     :  || ::  :    ::
3'           AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                  || : : :     : : : ::
3'                AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                                  || ::
3'                                AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'         ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
               :    : ||     :: :   :
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'  ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
      :  : ::  || : : : : ::  :: :  :
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
              ||       :      :       ::
3'            AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                                ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                                  || ::
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                              ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                                ||      ::
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                             ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                               ||   ::   ::
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                      ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                        : ||          :: :
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'     ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
             ||       : :       ::
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
    :      : ||  :       :  ::  :      :
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'        ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
            ||     :     : :     :     ::
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'    ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
       : ||          :     :          :: :
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'                          ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                             :    || ::    :
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'                       ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                              : || :
3' AGAGTCACCACAAGTTTAGTCAGCTGATGATTGCCA
```

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Bases

5'- ACC GTT AGT AGT CGA CTG ATT TGA ACA CCA CTG AGA -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na+ Conc: 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

Primary Sequence
5'- ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA -3'

Secondary Sequence
5'- TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG -3'

Maximum Delta G -69.79 kcal/mole

Delta G -8.13 kcal/mole
Base Pairs 5

```
5'                ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                  :       :  |||||   :       :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -6.68 kcal/mole
Base Pairs 3

```
5'                                    ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                                      |||
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -5.19 kcal/mole
Base Pairs 3

```
5'                          ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
```

```
                                    :    ::  |||
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                            ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                               ||
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -3.52 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                     :    |||  :    ::
3'            GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -3.52 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                   :       |||  :   :    :
3'               GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -3.3 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
           ::  :  :           |||      ::
3'        GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -3.17 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                        ::    :          |||
3'                     GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -3.17 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                          :          |||
3'                       GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5'           ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
               :  :  :   :   :: : :    ||
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -2.94 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
     : :     :       :       :: |||
3'   GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -2.94 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                |||       : :  ::    :
3'             GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -2.94 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                                  |||  :
3'                               GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -2.92 kcal/mole
Base Pairs 3

```
5'      ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
          :  :  :   |||   :  ::  :  :  :  ::
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5'                              ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                                : :    | |·|    : :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5'                            ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                                    : |||     : : :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
       |||       :  : :  : : :  :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
       : : |||   :   :    :    :    : :
3'    GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -2.3 kcal/mole
Base Pairs 3

```
5'                  ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                    :      |||          :   :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5'       ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
           :  : : :      ||          : :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.95 kcal/mole

Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
               :      :      ::    ||    :
3'           GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                                ||      : ::
3'                              GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                                  ||
3'                                GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
             :   : :  ::     || ::    :   ::
3'          GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5'                     ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                         : :           ||
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5'              ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                :      ||   :  ::    :     :  : :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5'          ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                 :  ||  :    :       : :     :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5'    ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
       :    : ||   :   :: :         ::
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                              :     ||
3'                            GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'                  ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                    :    :    : : :   :: ||
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'         ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
           :  :     :    :   :     :     : ||
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                   :  : : :   :  :  || :
3'                 GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
    :       :  ||                 :  :
3'  GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
         :      :   ||   ::     ::     :
3'       GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                          :  :     ||  :
3'                      GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
               ||   :      :
3'      GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                           : :: :  ||
3'                       GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                                ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                                  ||       :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'  ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
    :   :  : :    :      :  :  || :   :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                       ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                           ||  :          :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'               ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                      :   ||       :  :   :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                      ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                        ||   ::  : : :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'    ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
      ||                    :   ::
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                           ||         :
3'                       GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'                                  ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                                      :  ||    :
3'  GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

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Bases

5'- ACC GTT AGT AGT CGA CTG ATT TGA ACA CCA CTG AGA -3'

Target Type DNA

Oligo Conc 0.25 µM

Na+ Conc 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

Primary Sequence
5'- ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA -3'

Secondary Sequence
5'- TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT -3'

Maximum Delta G -67.39 kcal/mole

Delta G -8.13 kcal/mole
Base Pairs 5

```
5'            ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
              :   :   :   : |||||   :       :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -6.68 kcal/mole
Base Pairs 3

```
5'                                    ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                                      |||
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -5.24 kcal/mole
Base Pairs 4

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
```

```
                              ||||
3'                           TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -5.19 kcal/mole
Base Pairs 3

```
5'                            ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                                :    ::  |||
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -5 kcal/mole
Base Pairs 4

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
         :     :  : ||||  :    :: :::
3'        TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                          ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                              ||
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                    ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                        ||   :          :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'        ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
              :  ::: ||   :: : :    ::
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.53 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                  : :   |||    :
3'             TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.53 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                           :     : ||| :
3'                        TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -2.92 kcal/mole
Base Pairs 3

```
5'        ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
            : :   :  |||   :    :  : :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5'                      ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                        : :   |||   : :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5'                    ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                           : |||    : : :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5'          ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
            :  : |||  :       :  :   :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -2.3 kcal/mole
Base Pairs 3

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
          |||  :       :        .: :
3'   TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                     :  ||  :         : :
3'                    TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                                :     ||   :
3'                              TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                                : ||    :
3'                              TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                    .      :  ||     :     : :
3'                   TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                               ||          : :
3'                             TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole

Base Pairs 2

```
5'                ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                     :            ||
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
         :       ::  :             ||
3'    TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
          :   :       : :     :    ||
3'         TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                           :          ||
3'                     TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'               ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                 :  :    :  : : :   ::  ||
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
   :    :   :     :        ||  : :  :  ::
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                :   : :   :  :    ||
3'           TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                ::   :   : :       ||
3'              TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'   ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
     ::   :: :  : : : ||   :     : ::
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
           :       :     ||  :: :   :
3'    TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                :      :         : ||
3'              TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
         :  :   :  : ||   :     ::    :
3'          TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                :   || : :  :      :
3'              TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                          ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                            ||       :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'    ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
        || :  :   :          : :     :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
        :   ||      :   : ::  :
3'     TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                          : ||   : :
3'                        TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5' ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                                ||    :
3'                              TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'                              ACCGTTAGTAGTCGACTGATTTGAACACCACTGAGA
                                  :  ||    :
3'  TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

For questions regarding the Dimer Analysis contact our Technical Support Group
1-800-328-2661 or e-mail TechSupport@idtdna.com

Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- TCG GAT AGC TAG TCG TAC ATG TCT TTT CTT CCC TAG TAT G -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na^+ Conc: 50 mM

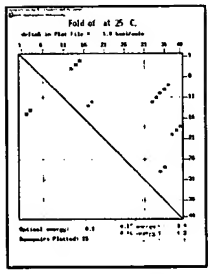
ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

mFold Input

General Information



CLICK TO VIEW DOT PLOT

Sequence Name: 40 Base Oligo
Batch Date: 11/8/2006
Sequence :
TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG

ADD TO ORDER

Structure 1

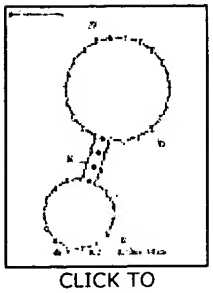
CLICK TO ENLARGE

ΔG	0.16 kcal.mole^{-1}
T_M	22.4 °C
ΔH	-22.7 kcal.mole^{-1}
ΔS	-76.8 cal.K^{-1}mole^{-1}

CONNECTIVITY | DETAILS

Structure 2

ΔG	0.25 kcal.mole^{-1}



CLICK TO ENLARGE

T_M	22.5	°C
ΔH	-34.8	kcal.mole^{-1}
ΔS	-117.7	cal.K^{-1}mole^{-1}

CONNECTIVITY DETAILS

Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- TCG GAT AGC TAG TCG TAC ATG TCT TTT CTT CCC TAG TAT G -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na+ Conc: 50 mM

ANALYZE | HAIRPIN | SELF-DIMER | HETERO-DIMER | NCBI BLAST | TM MISMATCH | LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

HOMO-DIMER ANALYSIS

Dimer Sequence
5'- TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG -3'

Maximum Delta G -69.79 kcal/mole

Delta G -8.26 kcal/mole
Base Pairs 6

```
5'                          TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                             ::  ||||||  ::
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -8.07 kcal/mole
Base Pairs 6

```
5'   TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
     :   :  :        ||||||        :  :   :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -4.64 kcal/mole
Base Pairs 3

```
5'        TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
            |||      ::  :  :   :  :  ::      :::
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -4.16 kcal/mole
Base Pairs 4

```
5'                          TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                              : :    ||||    : :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -4.16 kcal/mole
Base Pairs 4

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
           ||||       :       :       ::::
3'      GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -4.16 kcal/mole
Base Pairs 4

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                                      |||.|
3'                                GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -3.65 kcal/mole
Base Pairs 4

```
5'           TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                   :    :    ||||    :    :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                                        TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                                           ||
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                     : :  :   ||   :  : :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -3.43 kcal/mole
Base Pairs 3

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                    |||                 :::
3'                  GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5'      TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
        : ||  ::  :              :  ::  :: :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
       |||  :     : :  : : .   :  :::
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -2.44 kcal/mole
Base Pairs 3

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
      |||    :    : : : : : :    :    :::
3'    GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -2.3 kcal/mole
Base Pairs 3

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                : |||  :            :  ::: :
3'              GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5'          . TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
              : : ||  :         :  :: : :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5'  TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
          :  ||               ::  :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                    ||          ::
3'                GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                          : ||      :: :
3'                        GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'                                    TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                                      || ::
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'                          TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                            :  ||       ::  :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'              TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                  || :  ::    ::  : ::
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'            TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                || :      :      :      : : :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5'                                    TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                                        ||
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5'                   TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                       ||             : :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                                        ||
3'                                      GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'             TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                   :   :  ||   : :  :   :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'                                    TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                                        : || :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'                      TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                         :   ||         ::    :
3' GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
               :  ||                ::   :
3'            GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
           || : :   :          :   : : ::
3'         GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                                  : || :: :
3'                                GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                                       ||
3'                                   GTATGATCCCTTCTTTTCTGTACATGCTGATCGATAGGCT
```

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OligoAnalyzer 3.0

Bases

5'- TCG GAT AGC TAG TCG TAC ATG TCT TTT CTT CCC TAG TAT G -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na+ Conc: 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

Primary Sequence
5'- TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG -3'

Secondary Sequence
5'- TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT -3'

Maximum Delta G -69.79 kcal/mole

Delta G -8.26 kcal/mole
Base Pairs 6

```
5'                        TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                           ::  ||||||  ::
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -8.26 kcal/mole
Base Pairs 4

```
5'                     TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                        ||||   ::::   : :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -8.07 kcal/mole
Base Pairs 6

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
```

```
                    ||||||  :       :   :   :
3'                  TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -5.61 kcal/mole
Base Pairs 5

```
5'           TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
               |||||   : ::    :   : :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -4.64 kcal/mole
Base Pairs 3

```
5'   TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
      :   ::  : :   :   : : ::     |||
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -4.16 kcal/mole
Base Pairs 4

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
             :   :::    :          ||||
3'          TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                                  TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                                     ||
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'      TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
          : :  : ::: || ::   :  . :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.43 kcal/mole
Base Pairs 3

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
             :       |||         :       :
3'           TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.43 kcal/mole
Base Pairs 3

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                                    : :   |||
3'                                  TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.43 kcal/mole
Base Pairs 3

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                                      :  |||
3'                                    TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.14 kcal/mole
Base Pairs 2

```
5'          TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
              :     ||
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5'  TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
            :  :  :          :  ::  ||  :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5'              TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                : ||     : : : :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
      : :  :       : : :         :  |||
3'      TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                              :       |||   :
3'                          TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -2.44 kcal/mole
Base Pairs 3

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
               :   :                :    |||
3'          TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -2.3 kcal/mole
Base Pairs 3

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                                    :  |||
3'                                TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5'        TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
               :             :  || : :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
      :          :             ||  :
3'    TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole

Base Pairs 2

```
5'                                    TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                                      || ::
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'            TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
              :   :      :    ::  : ||
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'       TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
         :   ::      :  :   :      : ||
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'                      TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                          ||       ::  :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5'                                 TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                                      ||
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5'               TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                   :   :          ||
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
          :   :         ||         :   :
3'         TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                                      :   ||
3'                                TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
       ||     :      :        :   :   :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'          TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
            :           :     ||   :    :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
          : :  ||  :     : :           :   :
3'        TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'                              TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                                   :  ||  :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'                  TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                      :   :          ||   :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                        :  :        : : ||
3'              TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                                 :    : || :
3'                            TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
      : :  || :  ::   :         :  :   :
3'   TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTACATGTCTTTTCTTCCCTAGTATG
                                    || :
3'                               TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

For questions regarding the Dimer Analysis contact our Technical Support Group
1-800-328-2661 or e-mail TechSupport@idtdna.com

Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- TCG GAT AGC TAG TCG TCC GCC TAT CAT TAC ATG TTT -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na^+ Conc: 50 mM

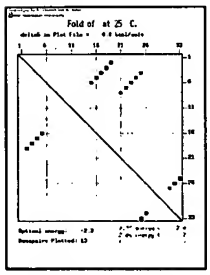
ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

mFold Input

General Information

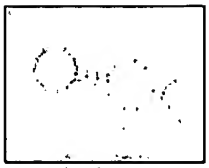


CLICK TO VIEW DOT PLOT

Sequence Name: 36 Base Oligo
Batch Date: 11/8/2006
Sequence :
TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT

ADD TO ORDER

Structure 1



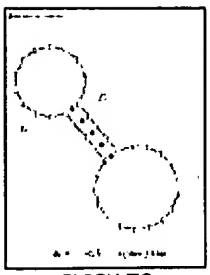
CLICK TO ENLARGE

ΔG	-2.31 kcal.mole^{-1}
T_M	37.7 °C
ΔH	-56.4 kcal.mole^{-1}
ΔS	-181.5 cal.K^{-1}mole^{-1}

CONNECTIVITY | DETAILS

Structure 2

ΔG	-1.47 kcal.mole^{-1}



CLICK TO ENLARGE

T_M	35.9	°C
ΔH	-42.7	kcal.mole^{-1}
ΔS	-138.2	cal.K^{-1}mole^{-1}

CONNECTIVITY DETAILS

Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- TCG GAT AGC TAG TCG TCC GCC TAT CAT TAC ATG TTT -3'

Target Type DNA
Oligo Conc 0.25 µM
Na^+ Conc 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE
ADD TO ORDER
DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

HOMO-DIMER ANALYSIS

Dimer Sequence
5'- TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT -3'

Maximum Delta G -67.39 kcal/mole

Delta G -8.26 kcal/mole
Base Pairs 6

```
5'                        TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
                           ::  ||||||  ::
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -8.26 kcal/mole
Base Pairs 4

```
5'                    TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
                      ||||   ::::   ::::
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -8.07 kcal/mole
Base Pairs 6

```
5' TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
                              ||||||
3'                        TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -5.61 kcal/mole
Base Pairs 5

```
5'          TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
              |||||    : :: :    :::::
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                                    TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
                                       ||
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'      TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
          : :   : ::: ||  :: ::: :   : :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
      ::      :      ||      :      ::
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.43 kcal/mole
Base Pairs 3

```
5' TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
                          : |||    ::: :
3'                       TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.14 kcal/mole
Base Pairs 2

```
5'           TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
               :    ||          ::     :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.14 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
       : :  :: :  :  ||  :  : ::  : :
3'   TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5'               TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
                 : ||     : : : :     :: :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'                                  TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
                                    || ::
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'                      TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
                          ||       ::
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5'                                TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
                                    ||
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
            :  : :       ||       : :  :
3'            TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5'       TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
         :   ||      :  : :  :      ::    :
3' TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
                    :      || ::      :
3'                TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
                       : : : || : : :
3'                   TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
                           ||       ::
3'                      TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
         : :  ||  :          :  ::  : :
3'       TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
             :   ||      ::      ::    :
3'          TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'                              TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
                                    :  ||  :
3'  TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'    TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
       :    ||    :    :     :    :   ::    :
3'  TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'  TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
                       :    ||     ::    :
3'                  TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'  TCGGATAGCTAGTCGTCCGCCTATCATTACATGTTT
                             :     ||     :
3'                         TTTGTACATTACTATCCGCCTGCTGATCGATAGGCT
```

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EXHIBIT B

Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- TAG TCG ACG ACC GTT AGG GTT TAC CTT CTT TGG GCC -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na+ Conc: 50 mM

ANALYZE | HAIRPIN | SELF-DIMER | HETERO-DIMER | NCBI BLAST | TM MISMATCH | LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

DILUTION | RESUSPENSION

RESULTS

SEQUENCE:
5'- TAG TCG ACG ACC GTT AGG GTT TAC CTT CTT TGG GCC -3'

COMPLEMENT:
5'- GGC CCA AAG AAG GTA AAC CCT AAC GGT CGT CGA CTA -3'

LENGTH:	36
GC CONTENT:	52.8 %
MELT TEMP:	66.6 °C
MOLECULAR WEIGHT:	11049.2 g/mole
EXTINCTION COEFFICIENT:	331700 L/(mole·cm)
nmole/OD_{260}:	3.01
µg/OD_{260}:	33.31

MELTING TEMPERATURE SETTINGS

TARGET TYPE:	DNA
OLIGO CONC	0.25 µM
Na^+ CONC	50 mM monovalent salt

MELTING TEMPERATURE ASSUMPTIONS AND LIMITATIONS

- Predictions are accurate for oligos from 8 to 60 bases in length, in neutral buffered solutions (pH 7 - 8) with monovalent cation concentrations from 10mM to 1.2 M.
- Oligo concentration is assumed to be significantly larger (at least 6x) than concentration of the complementary target, which is true in majority of molecular biology experiments. If this is not a case, concentration of the target cannot be ignored and you should enter in the box,

  Oligo Conc = [strand1] – [strand2]/2 when [strand1] ⩾ [strand2]
  Oligo Conc = ([strand1] + [strand2])/4 when [strand1] = [strand2]

- Melting temperature accuracy and models: (Oligo/Template)

  DNA/DNA: +/- 1.4°C (Allawi '97)
  LNA/DNA: +/- 2.0°C (McTigue '04)
  RNA/DNA: +/- 2.7°C (Sugimoto '95)
  DNA/RNA: +/- 2.7°C (Sugimoto '95)
  RNA/RNA: +/- 1.3°C (Xia '98)

Monovalent cation correction: +/- 2.0°C (Owczarzy '04)

- Non-consecutive LNA bases hybridized to a DNA template use a model from McTigue '04. Consecutive LNA bases on a DNA template and any LNA bases on an RNA template assume RNA energetic parameters and predictions are therefore less accurate.

- Effects of chemical modifications are neglected except when the modification contains a base, e.g., 5-Methyl dC, Internal Fluorescein dT. Energetic effects of these modifications are only approximated.

- This OligoAnalyzer version does not account for effects of divalent cations.

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- GGG TTT ACC TTC TTT GGG CC -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na^+ Conc: 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

DILUTION | RESUSPENSION

RESULTS

SEQUENCE:

5'- GGG TTT ACC TTC TTT GGG CC -3'

COMPLEMENT:

5'- GGC CCA AAG AAG GTA AAC CC -3'

LENGTH:	20
GC CONTENT:	55.0 %
MELT TEMP:	56.2 °C
MOLECULAR WEIGHT:	6106.0 g/mole
EXTINCTION COEFFICIENT:	175300 L/(mole·cm)
nmole/OD_{260}:	5.70
µg/OD_{260}:	34.83

MELTING TEMPERATURE SETTINGS

TARGET TYPE:	DNA
OLIGO CONC	0.25 µM
Na^+ CONC	50 mM monovalent salt

MELTING TEMPERATURE ASSUMPTIONS AND LIMITATIONS

- Predictions are accurate for oligos from 8 to 60 bases in length, in neutral buffered solutions (pH 7 - 8) with monovalent cation concentrations from 10mM to 1.2 M.
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- Melting temperature accuracy and models: (Oligo/Template)

  DNA/DNA: +/- 1.4°C (Allawi '97)
  LNA/DNA: +/- 2.0°C (McTigue '04)
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Monovalent cation correction: +/- 2.0°C (Owczarzy '04)

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Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- TCG GAT AGC TAG TCG TTG GGC TTT GAA GCC AGG AG -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na^+ Conc: 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

DILUTION | RESUSPENSION

RESULTS

SEQUENCE:
5'- TCG GAT AGC TAG TCG TTG GGC TTT GAA GCC AGG AG -3'

COMPLEMENT:
5'- CTC CTG GCT TCA AAG CCC AAC GAC TAG CTA TCC GA -3'

LENGTH:	35
GC CONTENT:	54.3 %
MELT TEMP:	67.0 °C
MOLECULAR WEIGHT:	10883.1 g/mole
EXTINCTION COEFFICIENT:	340000 L/(mole·cm)
nmole/OD_{260}:	2.94
µg/OD_{260}:	32.01

MELTING TEMPERATURE SETTINGS

TARGET TYPE:	DNA
OLIGO CONC	0.25 µM
Na^+ CONC	50 mM monovalent salt

MELTING TEMPERATURE ASSUMPTIONS AND LIMITATIONS

- Predictions are accurate for oligos from 8 to 60 bases in length, in neutral buffered solutions (pH 7 - 8) with monovalent cation concentrations from 10mM to 1.2 M.
- Oligo concentration is assumed to be significantly larger (at least 6x) than concentration of the complementary target, which is true in majority of molecular biology experiments. If this is not a case, concentration of the target cannot be ignored and you should enter in the box,

  Oligo Conc = [strand1] - [strand2]/2 when [strand1] ⩾ [strand2]
  Oligo Conc = ([strand1] + [strand2])/4 when [strand1] = [strand2]

- Melting temperature accuracy and models: (Oligo/Template)

  DNA/DNA: +/- 1.4°C (Allawi '97)
  LNA/DNA: +/- 2.0°C (McTigue '04)
  RNA/DNA: +/- 2.7°C (Sugimoto '95)
  DNA/RNA: +/- 2.7°C (Sugimoto '95)
  RNA/RNA: +/- 1.3°C (Xia '98)

Monovalent cation correction: +/- 2.0°C (Owczarzy '04)

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Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- TGG GCT TTG AAG CCA GGA G -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na^+ Conc: 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

DILUTION | RESUSPENSION

RESULTS

SEQUENCE:

5'- TGG GCT TTG AAG CCA GGA G -3'

COMPLEMENT:

5'- CTC CTG GCT TCA AAG CCC A -3'

LENGTH:	19
GC CONTENT:	57.9 %
MELT TEMP:	57.8 °C
MOLECULAR WEIGHT:	5908.9 g/mole
EXTINCTION COEFFICIENT:	185000 L/(mole·cm)
nmole/OD_{260}:	5.41
µg/OD_{260}:	31.94

MELTING TEMPERATURE SETTINGS

TARGET TYPE:	DNA
OLIGO CONC	0.25 µM
Na^+ CONC	50 mM monovalent salt

MELTING TEMPERATURE ASSUMPTIONS AND LIMITATIONS

- Predictions are accurate for oligos from 8 to 60 bases in length, in neutral buffered solutions (pH 7 - 8) with monovalent cation concentrations from 10mM to 1.2 M.
- Oligo concentration is assumed to be significantly larger (at least 6x) than concentration of the complementary target, which is true in majority of molecular biology experiments. If this is not a case, concentration of the target cannot be ignored and you should enter in the box,

  Oligo Conc = [strand1] – [strand2]/2 when [strand1] ⩾ [strand2]
  Oligo Conc = ([strand1] + [strand2])/4 when [strand1] = [strand2]

- Melting temperature accuracy and models: (Oligo/Template)

  DNA/DNA: +/- 1.4°C (Allawi '97)
  LNA/DNA: +/- 2.0°C (McTigue '04)
  RNA/DNA: +/- 2.7°C (Sugimoto '95)
  DNA/RNA: +/- 2.7°C (Sugimoto '95)
  RNA/RNA: +/- 1.3°C (Xia '98)

Monovalent cation correction: +/- 2.0°C (Owczarzy '04)

- Non-consecutive LNA bases hybridized to a DNA template use a model from McTigue '04. Consecutive LNA bases on a DNA template and any LNA bases on an RNA template assume RNA energetic parameters and predictions are therefore less accurate.
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Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- TAG TCG ACG ACC GTT ACA GCA TCC AAA AAC AAT TAG G -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na+ Conc: 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

DILUTION | RESUSPENSION

RESULTS

SEQUENCE:

5'- TAG TCG ACG ACC GTT ACA GCA TCC AAA AAC AAT TAG G -3'

COMPLEMENT:

5'- CCT AAT TGT TTT TGG ATG CTG TAA CGG TCG TCG ACT A -3'

LENGTH:	37
GC CONTENT:	43.2 %
MELT TEMP:	63.3 °C
MOLECULAR WEIGHT:	11359.4 g/mole
EXTINCTION COEFFICIENT:	372800 L/(mole·cm)
nmole/OD_{260}:	2.68
µg/OD_{260}:	30.47

MELTING TEMPERATURE SETTINGS

TARGET TYPE:	DNA
OLIGO CONC	0.25 µM
Na^+ CONC	50 mM monovalent salt

MELTING TEMPERATURE ASSUMPTIONS AND LIMITATIONS

- Predictions are accurate for oligos from 8 to 60 bases in length, in neutral buffered solutions (pH 7 - 8) with monovalent cation concentrations from 10mM to 1.2 M.
- Oligo concentration is assumed to be significantly larger (at least 6x) than concentration of the complementary target, which is true in majority of molecular biology experiments. If this is not a case, concentration of the target cannot be ignored and you should enter in the box,

  Oligo Conc = [strand1] – [strand2]/2 when [strand1] ⩾ [strand2]
  Oligo Conc = ([strand1] + [strand2])/4 when [strand1] = [strand2]

- Melting temperature accuracy and models: (Oligo/Template)

  DNA/DNA: +/- 1.4°C (Allawi '97)
  LNA/DNA: +/- 2.0°C (McTigue '04)
  RNA/DNA: +/- 2.7°C (Sugimoto '95)
  DNA/RNA: +/- 2.7°C (Sugimoto '95)
  RNA/RNA: +/- 1.3°C (Xia '98)

Monovalent cation correction: +/- 2.0°C (Owczarzy '04)

- Non-consecutive LNA bases hybridized to a DNA template use a model from McTigue '04. Consecutive LNA bases on a DNA template and any LNA bases on an RNA template assume RNA energetic parameters and predictions are therefore less accurate.
- Effects of chemical modifications are neglected except when the modification contains a base, e.g., 5-Methyl dC, Internal Fluorescein dT. Energetic effects of these modifications are only approximated.
- This OligoAnalyzer version does not account for effects of divalent cations.

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- CAG CAT CCA AAA ACA ATT AGG -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na+ Conc: 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

DILUTION | RESUSPENSION

RESULTS

SEQUENCE:
5'- CAG CAT CCA AAA ACA ATT AGG -3'
COMPLEMENT:
5'- CCT AAT TGT TTT TGG ATG CTG -3'

LENGTH:	21
GC CONTENT:	38.1 %
MELT TEMP:	50.1 °C
MOLECULAR WEIGHT:	6416.3 g/mole
EXTINCTION COEFFICIENT:	216100 L/(mole·cm)
nmole/OD_{260}:	4.63
µg/OD_{260}:	29.69

MELTING TEMPERATURE SETTINGS

TARGET TYPE:	DNA
OLIGO CONC	0.25 µM
Na^+ CONC	50 mM monovalent salt

MELTING TEMPERATURE ASSUMPTIONS AND LIMITATIONS

- Predictions are accurate for oligos from 8 to 60 bases in length, in neutral buffered solutions (pH 7 - 8) with monovalent cation concentrations from 10mM to 1.2 M.
- Oligo concentration is assumed to be significantly larger (at least 6x) than concentration of the complementary target, which is true in majority of molecular biology experiments. If this is not a case, concentration of the target cannot be ignored and you should enter in the box,

  Oligo Conc = [strand1] – [strand2]/2 when [strand1] ≥ [strand2]
  Oligo Conc = ([strand1] + [strand2])/4 when [strand1] = [strand2]

- Melting temperature accuracy and models: (Oligo/Template)

  DNA/DNA: +/- 1.4°C (Allawi '97)
  LNA/DNA: +/- 2.0°C (McTigue '04)
  RNA/DNA: +/- 2.7°C (Sugimoto '95)
  DNA/RNA: +/- 2.7°C (Sugimoto '95)
  RNA/RNA: +/- 1.3°C (Xia '98)

Monovalent cation correction: +/- 2.0°C (Owczarzy '04)

- Non-consecutive LNA bases hybridized to a DNA template use a model from McTigue '04. Consecutive LNA bases on a DNA template and any LNA bases on an RNA template assume RNA energetic parameters and predictions are therefore less accurate.
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- This OligoAnalyzer version does not account for effects of divalent cations.

Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- TCG GAT AGC TAG TCG TGA ATG GTT TTA TAG GAA CGC TA -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na^+ Conc: 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

DILUTION | RESUSPENSION

RESULTS

SEQUENCE:

5'- TCG GAT AGC TAG TCG TGA ATG GTT TTA TAG GAA CGC TA -3'

COMPLEMENT:

5'- TAG CGT TCC TAT AAA ACC ATT CAC GAC TAG CTA TCC GA -3'

LENGTH:	38
GC CONTENT:	42.1 %
MELT TEMP:	63.0 °C
MOLECULAR WEIGHT:	11787.7 g/mole
EXTINCTION COEFFICIENT:	379400 L/(mole·cm)
nmole/OD_{260}:	2.64
µg/OD_{260}:	31.07

MELTING TEMPERATURE SETTINGS

TARGET TYPE:	DNA
OLIGO CONC	0.25 µM
Na^+ CONC	50 mM monovalent salt

MELTING TEMPERATURE ASSUMPTIONS AND LIMITATIONS

- Predictions are accurate for oligos from 8 to 60 bases in length, in neutral buffered solutions (pH 7 - 8) with monovalent cation concentrations from 10mM to 1.2 M.
- Oligo concentration is assumed to be significantly larger (at least 6x) than concentration of the complementary target, which is true in majority of molecular biology experiments. If this is not a case, concentration of the target cannot be ignored and you should enter in the box,

  Oligo Conc = [strand1] – [strand2]/2 when [strand1] ⩾ [strand2]
  Oligo Conc = ([strand1] + [strand2])/4 when [strand1] = [strand2]

- Melting temperature accuracy and models: (Oligo/Template)

  DNA/DNA: +/- 1.4°C (Allawi '97)
  LNA/DNA: +/- 2.0°C (McTigue '04)
  RNA/DNA: +/- 2.7°C (Sugimoto '95)
  DNA/RNA: +/- 2.7°C (Sugimoto '95)
  RNA/RNA: +/- 1.3°C (Xia '98)

Monovalent cation correction: +/- 2.0°C (Owczarzy '04)

- Non-consecutive LNA bases hybridized to a DNA template use a model from McTigue '04. Consecutive LNA bases on a DNA template and any LNA bases on an RNA template assume RNA energetic parameters and predictions are therefore less accurate.

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Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- GAA TGG TTT TAT AGG AAC GCT A -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na^+ Conc: 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

DILUTION | RESUSPENSION

RESULTS

SEQUENCE:
5'- GAA TGG TTT TAT AGG AAC GCT A -3'
COMPLEMENT:
5'- TAG CGT TCC TAT AAA ACC ATT C -3'

LENGTH:	22
GC CONTENT:	36.4 %
MELT TEMP:	50.3 °C
MOLECULAR WEIGHT:	6813.5 g/mole
EXTINCTION COEFFICIENT:	225000 L/(mole·cm)
nmole/OD_{260}:	4.44
µg/OD_{260}:	30.28

MELTING TEMPERATURE SETTINGS

TARGET TYPE:	DNA
OLIGO CONC	0.25 µM
Na^+ CONC	50 mM monovalent salt

MELTING TEMPERATURE ASSUMPTIONS AND LIMITATIONS

- Predictions are accurate for oligos from 8 to 60 bases in length, in neutral buffered solutions (pH 7 - 8) with monovalent cation concentrations from 10mM to 1.2 M.
- Oligo concentration is assumed to be significantly larger (at least 6x) than concentration of the complementary target, which is true in majority of molecular biology experiments. If this is not a case, concentration of the target cannot be ignored and you should enter in the box,

  Oligo Conc = [strand1] - [strand2]/2 when [strand1] ⩾ [strand2]
  Oligo Conc = ([strand1] + [strand2])/4 when [strand1] = [strand2]

- Melting temperature accuracy and models: (Oligo/Template)

  DNA/DNA: +/- 1.4°C (Allawi '97)
  LNA/DNA: +/- 2.0°C (McTigue '04)
  RNA/DNA: +/- 2.7°C (Sugimoto '95)
  DNA/RNA: +/- 2.7°C (Sugimoto '95)
  RNA/RNA: +/- 1.3°C (Xia '98)

Monovalent cation correction: +/- 2.0°C (Owczarzy '04)

- Non-consecutive LNA bases hybridized to a DNA template use a model from McTigue '04. Consecutive LNA bases on a DNA template and any LNA bases on an RNA template assume RNA energetic parameters and predictions are therefore less accurate.
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- This OligoAnalyzer version does not account for effects of divalent cations.

Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- TAG TCG ACG ACC GTT AGG GTT TAC CTT CTT TGG GCC -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na^+ Conc: 50 mM

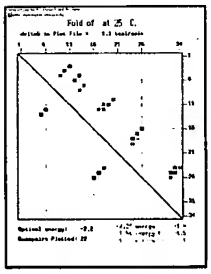
ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

mFold Input

General Information

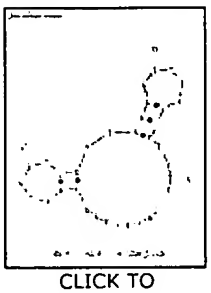


CLICK TO VIEW DOT PLOT

Sequence Name: 36 Base Oligo
Batch Date: 11/8/2006
Sequence :
TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC

ADD TO ORDER

Structure 1



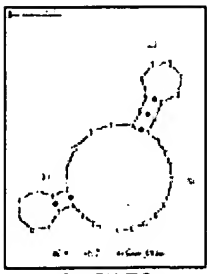
CLICK TO ENLARGE

ΔG -2.28 kcal.mole^{-1}
T_M 40.2 °C
ΔH -47.5 kcal.mole^{-1}
ΔS -151.6 cal.K^{-1}mole^{-1}

CONNECTIVITY | DETAILS

Structure 2

ΔG -1.73 kcal.mole^{-1}



CLICK TO ENLARGE

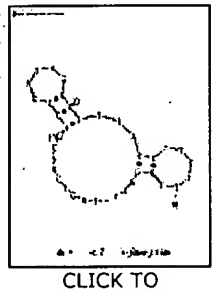
T_M 36.7 °C

ΔH -45.2 kcal.mole^{-1}

ΔS -145.9 cal.K^{-1}mole^{-1}

CONNECTIVITY DETAILS

Structure 3



CLICK TO ENLARGE

ΔG -1.66 kcal.mole^{-1}

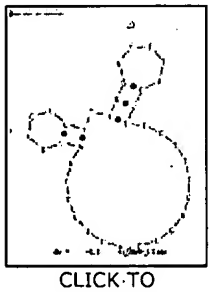
T_M 37.7 °C

ΔH -41.7 kcal.mole^{-1}

ΔS -134.2 cal.K^{-1}mole^{-1}

CONNECTIVITY DETAILS

Structure 4



CLICK TO ENLARGE

ΔG -1.15 kcal.mole^{-1}

T_M 33.7 °C

ΔH -38.8 kcal.mole^{-1}

ΔS -126.4 cal.K^{-1}mole^{-1}

CONNECTIVITY DETAILS

Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- TAG TCG ACG ACC GTT AGG GTT TAC CTT CTT TGG GCC -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na+ Conc: 50 mM

ANALYZE | HAIRPIN | SELF-DIMER | HETERO-DIMER | NCBI BLAST | TM MISMATCH | LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

HOMO-DIMER ANALYSIS

Dimer Sequence
5'- TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC -3'

Maximum Delta G -72.71 kcal/mole

Delta G -9.45 kcal/mole
Base Pairs 6

```
5'                              TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                : ||||||  :
3' CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -9.28 kcal/mole
Base Pairs 4

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                  ||||
3'                                CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -6.53 kcal/mole
Base Pairs 4

```
5'                          TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                            ||||  ::::
3' CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -4.95 kcal/mole
Base Pairs 3

```
5'                    TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                            |||   : : :
3' CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -4.67 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
       : :        |||      : : :        : :
3'       CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -4.41 kcal/mole
Base Pairs 3

```
5'          TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                :    ||| : : : : :    :
3' CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -4.41 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
             :     : |||  : : :  :     :
3'        CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                        TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                          : :    : || :    : :
3' CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                        TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                             ||      : :
3' CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'              TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                     : : || : :
3' CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5'           TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
              :    :   ||    ::   :    :
3' CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
           : ||       : : : :       :: :
3'          CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                          ||       ::
3'                       CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
           : ||    ::     ::     ::    :: :
3'          CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                          ||       ::
3'                       CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                  ||     :       :     ::
3'                 CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                   ||    :       :    ::
3'                  CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                  || ::
3'                                CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5'           TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
             ||    :    :      :    :    ::
3' CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5'         TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
           ||   :    : :  ::  : :    :   ::
3' CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'     TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
           || :                 : ::
3' CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
      : : ||  :: :    : ::  :: : :
3' CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'              TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                ::  :    || ::    :  ::
3' CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'           TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
               ||  ::          ::  ::
3' CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'                                    TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                      ||
3' CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
    : : :     : ||     :: :     : : :
3'  CCGGGTTTCTTCCATTTGGGATTGCCAGCAGCTGAT
```

For questions regarding the Dimer Analysis contact our Technical Support Group
1-800-328-2661 or e-mail TechSupport@idtdna.com

Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- TAG TCG ACG ACC GTT AGG GTT TAC CTT CTT TGG GCC -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na+ Conc: 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE
ADD TO ORDER
DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

Primary Sequence
5'- TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC -3'

Secondary Sequence
5'- TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGC -3'

Maximum Delta G -72.71 kcal/mole

Delta G -9.45 kcal/mole
Base Pairs 6

```
5'                              TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                : ||||||  :
3' CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -8.91 kcal/mole
Base Pairs 5

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                    : : :   : |||||
3'                  CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -6.53 kcal/mole
Base Pairs 4

```
5'                          TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
```

```
                                  ||||  ::::
3'  CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -5.23 kcal/mole
Base Pairs 4

```
5'  TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                 : :  ||||         ::   :
3'              CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -4.95 kcal/mole
Base Pairs 3

```
5'                     TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                            |||  :::
3'  CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -4.41 kcal/mole
Base Pairs 3

```
5'          TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
            :  : :    : :: : |||   :
3'  CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -3.9 kcal/mole
Base Pairs 3

```
5'  TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                               :  |||
3'                            CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -3.89 kcal/mole
Base Pairs 3

```
5'  TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
              :   : :  |||   : :: :
3'             CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -3.89 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
              :      : |||     :   :      :
3'            CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -3.89 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                          ::   :   ||| ::
3'                      CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -3.89 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                             :: |||   :
3'                       CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                         TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                           ::    : || :    ::
3' CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                         TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                              ||       ::
3' CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                    :      : || : :
3' CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -3.29 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
       :   :  |||   :  : :  :     :
3'       CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -3.14 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                   :    :   :: :    ||
3'                 CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -3.14 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                    ||
3'                                  CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5'           TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
             :  : :: :       ||   :    :
3' CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
          : :    :          ::     || :
3'        CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                :     :  :   : :   || :
3'          CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -3.07 kcal/mole

Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
      : :       :: || :        : : : :
3'    CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
               :: || :     :        : :
3'     CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                       ::     ::    ||
3'                CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
     :   :       || :     :        : :
3'   CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
               : ::     ||       :    :
3'             CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                     : :    || :: :    :
3'                   CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                              :  ||
3'                        CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
          :    ||    ::        :   :
3'         CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                      ||   ::      :
3'                 CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                             ::  ||
3'                        CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5'            TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
              :                 :   :   ||
3' CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5'          TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
              :       :  ::  : :   :  ||
3' CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                  ||     :
3'                                CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'      TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                          :        : ||
3' CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'  TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
              :   :  : ::  || : :
3' CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'        TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
          :  :    ||   :        :        :
3' CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'             TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
               : :     :           ||  ::
3' CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                 TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                   :  :    || ::    :  ::
3' CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                      TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                          ||    : :     :
3' CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'   TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
     : : : :       : :   ||      :  :
3' CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'       TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
         : ||    :   : :         : :     :
3' CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'                                      TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                        ||
3' CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                  : : : || :      : : :
3' CGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

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Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- TAG TCG ACG ACC GTT AGG GTT TAC CTT CTT TGG GCC -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na^+ Conc: 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

Primary Sequence
5'- TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC -3'

Secondary Sequence
5'- TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG -3'

Maximum Delta G -72.71 kcal/mole

Delta G -7.87 kcal/mole
Base Pairs 5

```
5'                TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                     : |||||
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -6.68 kcal/mole
Base Pairs 3

```
5'                          TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                             : : :     |||
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -6.53 kcal/mole
Base Pairs 4

```
5'                   TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
```

```
                          ||||  :  :   :   :
3'  GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -6.21 kcal/mole
Base Pairs 3

```
5'  TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                         :        :    |||
3'                     GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -6.21 kcal/mole
Base Pairs 3

```
5'  TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                 ::   |||
3'                               GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -5.19 kcal/mole
Base Pairs 3

```
5'                                TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                  ::  |||
3'  GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -5.19 kcal/mole
Base Pairs 3

```
5'                                TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                    :   |||
3'  GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -5.12 kcal/mole
Base Pairs 4

```
5'  TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                           :  ||||   :   :
3'                      GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -5.02 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                   |||
3'                               GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -4.67 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                             |||   : :
3'                          GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'           TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
             :    : : : ||    : : : :
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.54 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                        : :   |||     :
3'                       GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.14 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
          :  : :           : :  :     ||
3'         GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5'           TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
             :       :          : :  ||
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
          : :    : :    ::          ||
3'    GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5'        TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
            :: : : ||
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                      :       :      : ||
3'                   GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5'        TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
            :  :   ||     :  :      : :
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
              :     || :: :     :     : :
3'              GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                  :    || : :      :      : :
3'                GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole

Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
            ||           : :          :
3'          GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                    ||
3'                                  GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5'                          TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                            |||     :
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5'            TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
               :      : ::   |||    :
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
          :      ||                : :
3'       GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5'     TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
         : :  : :      :  :  :: ||
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
       : :  :            ::   || :  :
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
      :      :             || :
3'  GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
      :  :   : : :  :   : :  ||      :
3'     GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5'              TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                 || : :    :          :
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
     ::   :       ||          :   :   :
3'   GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                           || ::      :
3'                         GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'                                  TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                      ||
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'      TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
        : :       : :   : :  :  :  ||
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                              || : :   :
3'                            GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'                TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                : ||
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'          TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                 :   :    :      || :
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

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OligoAnalyzer 3.0

Bases

5'- TAG TCG ACG ACC GTT AGG GTT TAC CTT CTT TGG GCC -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na+ Conc: 50 mM

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HAIRPIN
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CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

Primary Sequence
5'- TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC -3'

Secondary Sequence
5'- TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA -3'

Maximum Delta G -72.71 kcal/mole

Delta G -7.87 kcal/mole
Base Pairs 5

```
5'                    TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                        :  |||||
3'  ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -6.9 kcal/mole
Base Pairs 4

```
5'  TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
              : ||||     :  :  :    :    :  :
3'            ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -6.68 kcal/mole
Base Pairs 3

```
5'                              TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
```

```
                                  : : :     |||
3'  ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -6.53 kcal/mole
Base Pairs 4

```
5'                          TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                            :   |||| : :   :   :
3'  ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -5.19 kcal/mole
Base Pairs 3

```
5'                                       TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                         :::  |||
3'  ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -5.19 kcal/mole
Base Pairs 3

```
5'                                     TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                       :   |||
3'  ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -4.67 kcal/mole
Base Pairs 3

```
5'  TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                         :  : |||
3'                     ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -4.41 kcal/mole
Base Pairs 3

```
5'          TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
            :  : . :  ||| ::   :  :: ::
3'  ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -4.41 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
             :  :         |||  :     :
3'          ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'               TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                  :  : :     ||     : : : :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
       ||          :     : :  :  : :       :
3'  ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
          ||  : :         :     :      : :
3'      ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.52 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
           :                 : |||
3'        ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.52 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                        :  : ||| :
3'                     ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.29 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                ::: |||     :          :
3'              ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.14 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                    ||
3'                                  ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5'            TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
              ::         :     :  :  ||
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
   :::     :        :    :: ::          ||
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                  :    ||
3'                    ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
      : :     ||  ::         :      : :
3'    ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole

Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                    :  ||
3'                                  ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5'                                  TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                    |||     :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5'                TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                   :  :    ::   |||    :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                :      : :      ||    : :
3'           ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                :       : ::     : ||
3'            ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                     :  ||  :   :
3'                 ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                  ||
3'                          ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                  : ||
3'                          ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5'    TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
        ::    :      :      ::    || :  :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5'   TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
       ::                        ||    :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                        :      : || :     :
3'                      ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'                                  TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                                      ||
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'          TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                  :    :  : : :  :  :  ||
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'                     TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                        : ||        : ::
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'           TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
             ::     || :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'            TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                    :   :   :      || :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
         :        ||          :          :
3'       ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                    :    ||             :
3'              ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTAGGGTTTACCTTCTTTGGGCC
                          || : :   :
3'                        ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

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Instructions | Definitions | Feedback

IDT SciTools OligoAnalyzer 3.0

Bases

5'- TAG TCG ACG ACC GTT ACA GCA TCC AAA AAC AAT TAG G -3'

Target Type: DNA

Oligo Conc: 0.25 µM

Na^+ Conc: 50 mM

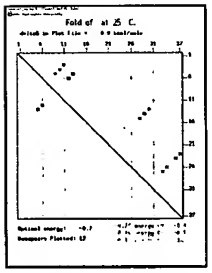
ANALYZE | HAIRPIN | SELF-DIMER | HETERO-DIMER | NCBI BLAST | TM MISMATCH | LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

mFold Input

General Information



CLICK TO VIEW DOT PLOT

Sequence Name: 37 Base Oligo

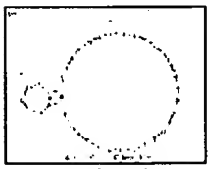
Batch Date: 11/8/2006

Sequence :

TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG

ADD TO ORDER

Structure 1



CLICK TO ENLARGE

ΔG	-0.72	kcal.mole^{-1}
T_M	34.4	°C
ΔH	-23	kcal.mole^{-1}
ΔS	-74.8	cal.K^{-1}mole^{-1}

CONNECTIVITY | DETAILS

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Bases

5'- TAG TCG ACG ACC GTT ACA GCA TCC AAA AAC AAT TAG G -3'

Target Type: DNA
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ANALYZE
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HETERO-DIMER
NCBI BLAST
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LNA CONVERSION

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RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

HOMO-DIMER ANALYSIS

Dimer Sequence
5'- TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG -3'

Maximum Delta G -69.59 kcal/mole

Delta G -9.45 kcal/mole
Base Pairs 6

```
5'                          TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                            : |||||| :
3' GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -6.53 kcal/mole
Base Pairs 4

```
5'                        TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                          |||| :::: 
3' GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -5.36 kcal/mole
Base Pairs 4

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                          ||||
3'                        GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -4.95 kcal/mole
Base Pairs 3

```
5'                    TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                            |||  :::
3' GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                          TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                            ::   : || :   ::
3' GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                          TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                               ||      ::
3' GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                 TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                      :      : || :      :
3' GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -3.29 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
             :  |||   : :: :   ::: :
3'       GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -3.14 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
      :    :    :    ||    :    :    :
3'  GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
           ||          :   :          ::
3'           GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                        || ::      :: ::
3'                      GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
         :       ||  :   :  ::      :
3'   GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
       : :       ||           ::       : :
3'     GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
       :         ||   :   :   ::         :
3'      GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
               ||                ::
3'          GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                          : ||       :: :
3'                       GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                          :  ||     ::  :
3'                         GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                               ||    ::
3'                             GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'          TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
            :  : || :           : :: :  :
3' GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'        TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
          :  :    ||   :   :   ::    :  :
3' GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                     :   ||          ::   :
3'                   GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                  TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                     :  :     || ::     :  :
3' GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'                      TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                         ||     : :     ::
3' GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'           TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
             :  : :     : || :: :     : :  :
3' GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'        TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
          : ||    :    : :  : :    :     :: :
3' GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'                                      TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                                        ||
3' GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'          TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
               :        :  ||  :        : *
3' GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'    TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
      || : :       : : : : : :       : : ::
3' GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                  ||      :      :      ::
3'              GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                                      ||
3'                                  GGATTAACAAAAACCTACGACATTGCCAGCAGCTGAT
```

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5'- TAG TCG ACG ACC GTT ACA GCA TCC AAA AAC AAT TAG G -3'

Target Type: DNA
Oligo Conc: 0.25 µM
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ANALYZE | HAIRPIN | SELF-DIMER | HETERO-DIMER | NCBI BLAST | TM MISMATCH | LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

Primary Sequence
5'- TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG -3'

Secondary Sequence
5'- TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG -3'

Maximum Delta G -70.77 kcal/mole

Delta G -7.87 kcal/mole
Base Pairs 5

```
5'                TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                     : |||||         :
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -6.97 kcal/mole
Base Pairs 4

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
              : : ::     ||||  :: :
3'        GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -6.68 kcal/mole
Base Pairs 3

```
5'                          TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
```

```
                              : : :     |||
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -6.53 kcal/mole
Base Pairs 4

```
5'                    TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                         ||||  : :   : :
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -6.12 kcal/mole
Base Pairs 4

```
5'            TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
              :   : : : ::        ||||  :
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -5.84 kcal/mole
Base Pairs 4

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                 : : :   : ||||  : :
3'               GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -5.19 kcal/mole
Base Pairs 3

```
5'                                TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                                  : :  |||
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -5.19 kcal/mole
Base Pairs 3

```
5'                                TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                                    :   |||
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -4.74 kcal/mole
Base Pairs 3

```
5'          TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
            :    :          |||   :  :
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -4.74 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
     :  :   : : : |||       :      :
3'     GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -4.64 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                          |||       :
3'                       GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.9 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                :    :: :: ::   |||  :
3'              GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.9 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                        :  :      |||
3'                      GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.89 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                      :        ||| :     :
3'                GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.89 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                          :   |||     : :
3'                  GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.29 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
              : :  :  ::      ||| : :::
3'          GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5'        TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
            :: : : ||                 :
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5'       TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
           :  :   ||      :  : : :
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
          :  : :        : || ::
3'         GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                                    ||
3'                                GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole

Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
            ||         :  :  ::  : :
3'          GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5'                          TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                            |||    :
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5'  TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
    :  : :          || :  : :   :
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
      ::   :       :   || : :  : : ::  :
3'   GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                                  ||
3'                  GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                   :  :   :  ||     :
3'                 GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
          :     ||  :    : :  :  :  ::
3'       GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                                    ||
3'                            GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5'               TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                 || : :     :      : :
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                            :          ||
3'                        GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'                                TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                                     ||
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'          TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                 :   :    ||    :      :
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'              TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                :       : ||    ::        : :
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'       TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
         : :        : :    ||  :    : :      :
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'                   TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                                 : ||     :
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

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RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

Primary Sequence
5'- TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG -3'

Secondary Sequence
5'- TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA -3'

Maximum Delta G -70.41 kcal/mole

Delta G -7.87 kcal/mole
Base Pairs 5

```
5'                  TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                      :  |||||         :
3'  ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -7.18 kcal/mole
Base Pairs 5

```
5'  TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                :  :         |||||  :::
3'              ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -6.9 kcal/mole
Base Pairs 4

```
5'  TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
```

```
            : |||| :    :       :   : :
3'          ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -6.68 kcal/mole
Base Pairs 3

```
5'                              TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                                 : : :     |||
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -6.53 kcal/mole
Base Pairs 4

```
5'                        TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                          :   |||| : :   : :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -6.12 kcal/mole
Base Pairs 4

```
5'              TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                 :  : :    ::       |||| :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -5.83 kcal/mole
Base Pairs 4

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                       : :  |||| :: : : :
3'              ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -5.19 kcal/mole
Base Pairs 3

```
5'                                    TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                                      ::  |||
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -5.19 kcal/mole
Base Pairs 3

```
5'                                  TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                                       :   |||
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -5.02 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
          :            : |||   :: : :::
3'        ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -4.74 kcal/mole
Base Pairs 3

```
5'             TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
               ::         :       |||   :  :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -4.74 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                    ||| : :  : : ::  :
3'                  ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -4.64 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                          ||| : :: :
3'                 ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -4.41 kcal/mole
Base Pairs 3

```
5'        TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
          :  :  :  ||| ::   :  :    :  : :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.89 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                    :    |||         :
3'            ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.89 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                :: :  : : |||   :
3'              ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
      ||          :     ::   :     :
3'  ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
         ||  : :   :         :
3'     ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.43 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
            :  : : : ||| ::  :  :  ::
3'   ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
      : :    ||  :: : :      :  :: :
3'    ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole

Base Pairs 3

```
5'                                  TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                                    |||     :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
   |||     :        ::       : :  :    :  :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                                    |||
3'                                  ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5'       TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
         :         : :  : ||  :       : :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5'    TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
        ::                ||      :   :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5'  TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
           : :       ||    : :  : : ::  :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                :        ::  ||  :
3'            ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                                 :  ||
3'                    ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                                 :  ||
3'                     ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                                   :   ||
3'                                ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'                                    TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                                        ||
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'                     TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                        : ||        : ::    :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'     TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
         ||     :       :        : :         :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                          :        : || :
3'                        ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'             TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                     :   :   ||     :       :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'               TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                  :   :     ||    : :       : :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'          TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                  :   :  : ||  :    : :     :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5'            TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
              : :     ||  :                   :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                                  :    ||
3'                          ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
         :       ||  :   :  :     :   :  :
3'       ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' TAGTCGACGACCGTTACAGCATCCAAAAACAATTAGG
                                 :        ||
3'                             ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

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Instructions | Definitions | Feedback

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OligoAnalyzer 3.0

Bases

5'- TCG GAT AGC TAG TCG TTG GGC TTT GAA GCC AGG AG -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na^+ Conc: 50 mM

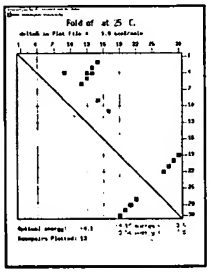
ANALYZE | HAIRPIN | SELF-DIMER | HETERO-DIMER | NCBI BLAST | TM MISMATCH | LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

mFold Input

General Information

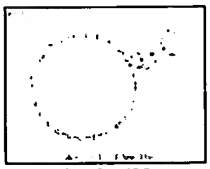


CLICK TO VIEW DOT PLOT

Sequence Name: 35 Base Oligo
Batch Date: 11/8/2006
Sequence :
TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG

ADD TO ORDER

Structure 1



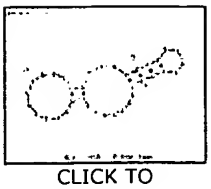
CLICK TO ENLARGE

ΔG -4.13 kcal.mole^{-1}
T_M 58.7 °C
ΔH -40.4 kcal.mole^{-1}
ΔS -121.8 cal.K^{-1}mole^{-1}

CONNECTIVITY | DETAILS

Structure 2

ΔG -3.97 kcal.mole^{-1}



CLICK TO ENLARGE

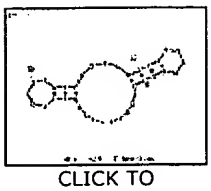
T_M 45.2 °C

ΔH -63.1 kcal.mole^{-1}

ΔS -198.2 cal.K^{-1}mole^{-1}

CONNECTIVITY DETAILS

Structure 3



CLICK TO ENLARGE

ΔG -2.91 kcal.mole^{-1}

T_M 40.6 °C

ΔH -58.2 kcal.mole^{-1}

ΔS -185.5 cal.K^{-1}mole^{-1}

CONNECTIVITY DETAILS

Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- TCG GAT AGC TAG TCG TTG GGC TTT GAA GCC AGG AG -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na^+ Conc: 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

HOMO-DIMER ANALYSIS

Dimer Sequence
5'- TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG -3'

Maximum Delta G -70.77 kcal/mole

Delta G -9.75 kcal/mole
Base Pairs 5

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                :    |||||  :::::    :
3'              GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -8.26 kcal/mole
Base Pairs 6

```
5'                      TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                         ::  ||||||  ::
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -5.02 kcal/mole
Base Pairs 3

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                   |||  :   :  :::
3'              GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -4.74 kcal/mole
Base Pairs 3

```
5'          TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
              : |||     ::     ::: :
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -4.74 kcal/mole
Base Pairs 3

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
    :    : |||                  ::: :    :
3'  GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -4.16 kcal/mole
Base Pairs 4

```
5'                  TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                     :  :   ||||   :  :
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                                TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                                   ||
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.14 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
          :: :        ||        : ::
3'       GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.14 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                          :    ||    :
3'                      GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5'     TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
         ||  :     ::  :     :  ::     :  ::
3'  GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5'  TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                                  ||  ::
3'                                GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5'  TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                             ||     ::
3'                        GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5'  TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                   ||          ::
3'          GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5'  TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                    :    :  ||  ::  :    :
3'                 GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5'  TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                          ||          ::
3'                     GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
          ||  : : :          : : :  ::
3'         GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                        ||  .         ::
3'                      GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'                                  TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                                    || ::
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'                     TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                         ||        ::
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'           TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
             ||  :   : :    : :   :  ::
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
               ||   :  :   ::
3'     GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'  TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
    ||         :              :         ::
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
              || : :           : : ::
3'            GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5'                              TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                                  ||
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'                             TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                                 : || :
3' GAGGACCGAAGTTTCGGGTTGCTGATCGATAGGCT
```

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Instructions | Definitions | Feedback

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OligoAnalyzer 3.0

Bases

5'- TCG GAT AGC TAG TCG TTG GGC TTT GAA GCC AGG AG -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na^+ Conc: 50 mM

ANALYZE
HAIRPIN
SELF-DIMER
HETERO-DIMER
NCBI BLAST
TM MISMATCH
LNA CONVERSION

CLEAR SEQUENCE
ADD TO ORDER
DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

Primary Sequence
5'- TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG -3'

Secondary Sequence
5'- TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA -3'

Maximum Delta G -70.77 kcal/mole

Delta G -8.26 kcal/mole
Base Pairs 6

```
5'                          TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                             ::  ||||||  ::
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -6.9 kcal/mole
Base Pairs 4

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                ||||    :   :         :
3'            ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -5.7 kcal/mole
Base Pairs 4

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
```

```
        ||||     :   :              :  :   ::
3'      ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -5.02 kcal/mole
Base Pairs 3

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
               :  :      ::  :  ::  |||
3'             ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -4.74 kcal/mole
Base Pairs 3

```
5'           TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                :     :    ::     |||  :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -4.74 kcal/mole
Base Pairs 3

```
5'   TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
      ::       ::                 |||  :    :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -4.74 kcal/mole
Base Pairs 3

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                                |||
3'                              ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -4.16 kcal/mole
Base Pairs 4

```
5'                        TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                             :   ||||    :   :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                                    TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                                      ||
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.14 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                      || : :       :
3'                   ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5'        TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
          : :  : : :            : : :     : ||
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.07 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                      :       ||    :
3'                    ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
   : :  :  |||              : :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
           |||    :
3'         ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.95 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                                :  ||  ::
3'          ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5'      TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
           ::   :  : ||       :         ::
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                          ||      : ::
3'    ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
       :  : :  :        : || : : :  :
3'     ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                    :      :  ||  :     :
3'              ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                      :  : :  ||  :
3'                ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole

Base Pairs 2

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                  : :  ||   :.  :
3'                ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                                .||
3'                              ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.6 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                                   ||
3'                                 ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'           TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                  :   :    : :   :  ||
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'                                 TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                                   || : :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'                      TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                          ||       : :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'                          TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                            ||
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'           TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
               : : : : ||          :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
              || :               :
3'        ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5'                                  TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                                      ||
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5'                 TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                     ||              :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
             ||   :         :  :  :
3'         ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'                                  TCGGATAGCTAGTCGTTGGGCTTTGAAGCCAGGAG
                                      :  ||  :
3'  ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

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5'- TCG GAT AGC TAG TCG TGA ATG GTT TTA TAG GAA CGC TA -3'

Target Type: DNA

Oligo Conc: 0.25 µM

Na^+ Conc: 50 mM

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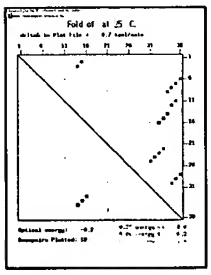


CLICK TO VIEW DOT PLOT

Sequence Name: 38 Base Oligo

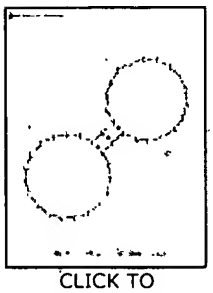
Batch Date: 11/8/2006

Sequence :

TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA

ADD TO ORDER

Structure 1



CLICK TO ENLARGE

ΔG -0.2 kcal.mole^{-1}

T_M 27.4 °C

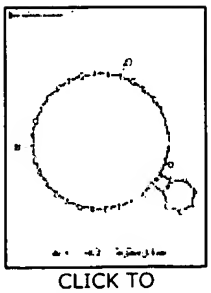
ΔH -25.1 kcal.mole^{-1}

ΔS -83.5 cal.K^{-1}mole^{-1}

CONNECTIVITY | DETAILS

Structure 2

ΔG -0.18 kcal.mole^{-1}



CLICK TO ENLARGE

T_M	28.3 °C
ΔH	-18.5 kcal.mole^{-1}
ΔS	-61.4 cal.K^{-1}mole^{-1}

CONNECTIVITY DETAILS

Instructions | Definitions | Feedback

IDT SciTools
OligoAnalyzer 3.0

Bases

5'- TCG GAT AGC TAG TCG TGA ATG GTT TTA TAG GAA CGC TA -3'

Target Type: DNA
Oligo Conc: 0.25 µM
Na^+ Conc: 50 mM

ANALYZE | HAIRPIN | SELF-DIMER | HETERO-DIMER | NCBI BLAST | TM MISMATCH | LNA CONVERSION

CLEAR SEQUENCE | ADD TO ORDER | DEFAULT SETTINGS

RESULTS | BASE NOTATION | 5' MODS | INTERNAL MODS | 3' MODS

HOMO-DIMER ANALYSIS

Dimer Sequence
5'- TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA -3'

Maximum Delta G -70.41 kcal/mole

Delta G -8.26 kcal/mole
Base Pairs 6

```
5'                        TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
                           ::. ||||||  ::
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -5.7 kcal/mole
Base Pairs 4

```
5' TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
       ||||    :   ::      ::   :    ::::
3'       ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -4.95 kcal/mole
Base Pairs 3

```
5' TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
                |||   :         :   :::
3'            ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -4.16 kcal/mole
Base Pairs 4

```
5'                    TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
                        :   ||||   :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'                                    TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
                                       ||
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'            TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
                :    :   ||   :    :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5'   TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
      ||      : :               : :      : :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.61 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
                                    : || :
3'                                  ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.4 kcal/mole
Base Pairs 4

```
5' TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
                    :  : :  ||||  : :  :
3'                  ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.29 kcal/mole
Base Pairs 3

```
5' TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
                    :  |||       :::  :
3'                  ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -3.14 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
                                    ||
3'                                  ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5' TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
   : :  :  |||       ::       :::  :  : :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5' TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
          |||   :                :   :::
3'          ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -2.56 kcal/mole
Base Pairs 3

```
5' TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
                                |||     :::
3'                              ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -2.44 kcal/mole
Base Pairs 3

```
5'        TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
          ::  |||              :::  ::
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
       :  : :  :     ||   ::     :  : :  :
3'    ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
            :     : ||    :: :      :
3'     ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
                     :  ||  ::  ::  :
3'                  ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.94 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
                       : :  ||     ::  : :
3'                     ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'                                    TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
                                      || ::
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'                          TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
                              ||       ::
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'                        TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
                          ||                 ::
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5'           TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
                 : : : : ||  :: : : : : :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.57 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
              || :  :       :  : ::
3'        ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5'                                  TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
                                       ||
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5'                  TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
                       ||              ::
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.47 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
                       ||       ::
3'           ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -1.34 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
              ||     :          :     ::
3'          ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'                              TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
                                   : || :
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
                    :  :    : || :    :  :
3'                 ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5'      TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
            ||    :  : :  : :  :    ::
3' ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
                              ||  :    :  ::
3'                            ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

Delta G -0.96 kcal/mole
Base Pairs 2

```
5' TCGGATAGCTAGTCGTGAATGGTTTTATAGGAACGCTA
                                        ||
3'                                      ATCGCAAGGATATTTTGGTAAGTGCTGATCGATAGGCT
```

For questions regarding the Dimer Analysis contact our Technical Support Group
1-800-328-2661 or e-mail TechSupport@idtdna.com